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SYMPOSIUM

The Blood Disorders of Early Infancy

INTRODUCTION

SPECIAL EDITOR
CARL H. SMITH, M.D.

THE subject matter of this symposium was chosen because of current interest in the manifold problems of the newborn period. This early period of life has been conspicuous for the intensive investigation of normal physiologic processes and of their aberrations. From the standpoint of hematology, this is the time when the blood picture may be obscured by the effects upon hematopoiesis of profound alterations in the other systems of the body following birth. In addition, the hematopoietic system itself undergoes concurrent modifications. Blood disorders encountered in this period must, therefore, be interpreted in the light of physiologic developments within the hematopoietic system as well as of those of other systems which affect it indirectly.

The search for a fetal etiology of congenital anomalies has had its impact on studies of the blood dyscrasias in the first months of life. This has prompted an investigation of conditioning factors which may be operative in, and antedating the perinatal period, to the critical first month of gestation. So impelling has been this orientation that the pathogenesis and interpretation of the blood disorders included within the scope of this symposium necessitate a thorough examination of maternal-fetal relationships and of environmental and genetic influences in conjunction with a consideration of normal embryological development. The problems of diagnosis and therapy hinge, therefore, on an understanding of the many interrelated mechanisms that play a role in the genesis of the various blood elements and in the synthesis of the coagulation factors. In the light of recent investigations, many of the abnormalities included in this symposium may eventually be included in the ever-growing list of inborn errors of metabolism which find their inception in the developing fetus.

This introduction would not be complete without an expression of gratitude to the individual authors who gave so liberally of their time and efforts to prepare outstanding contributions containing the newer knowledge and viewpoints. It is hoped that pediatricians will find these efforts both stimulating and rewarding.

THE ANTENATAL BACKGROUND OF BLOOD DISEASES OF INFANCY

EDITH L. POTTER, M.D., PH.D.
CHICAGO, ILL.

ABNORMALITIES of the blood having a prenatal origin may be discernible immediately after birth or may not be discovered until months or years later. Those most commonly presenting symptoms immediately after birth relate to a lack of red blood cells, although deficiencies of other constituents of blood such as platelets, fibrin, and globulin may also be demonstrable in the newborn period. Those with symptoms appearing later are for the most part inherited abnormalities of enzyme systems or of individual components of blood cells.

Anemia at birth may be the result of (1) deficient formation of red blood cells, (2) loss of them by hemorrhage, or (3) abnormal destruction of them. Hypochromic anemia in which the number of circulating erythrocytes is normal but in which they contain a decreased amount of hemoglobin is rare.

The total blood volume, the concentration of erythrocytes, and the hemoglobin which is normal or desirable for a newborn infant have been the subjects of mild disagreement among different investigators. It has been stated that when the cord is clamped immediately the infant is "deprived" of 100 c.c. or more of blood. On the other hand, since the volume of blood in the fetus and placenta is in equilibrium before birth, the addition of blood from the placenta to that normally present within the body of the fetus could be responsible for a temporary increase in volume which under some circumstances might be a detriment to the infant. The total volume of blood within the body of the fetus or infant at birth is generally given as approximately 100 c.c. per kilogram of body weight. Erythrocyte levels and hemoglobin concentration are reported to be slightly higher during early days of life in infants who have received placental blood than those with the cord clamped immediately after birth, but they are approximately the same by the end of the first month.

Studies on capillary blood of 244 normal full-term, 1-day-old infants born at the Chicago Lying-in Hospital[1] showed a mean hemoglobin of 20.7 Gm. with a range of from 15.2 to 26.2 Gm. and a mean erythrocyte count of 6.19 million cells with a range of from 4.33 to 8.05. By the eighth day, the mean hemoglobin had fallen to 18.4 Gm. and the erythrocytes to 5.47 million.

These figures are slightly higher than the averages given by Smith,[2] which he obtained by averaging data from several reports; these averages were 5.64 million erythrocytes and 19.85 Gm. of hemoglobin on the first

From the Department of Obstetrics and Gynecology, The University of Chicago, and the Chicago Lying-in Hospital.

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day of life. Venous blood invariably has a lower concentration of cells; in the averages obtained by Smith, the erythrocytes were 4.80 million and hemoglobin 17.0 Gm. on cord blood or venous blood obtained during the first day. Walker and Turnbull[3] reported that hemoglobin levels rise after the thirty-sixth week, with a mean level at 40 weeks of gestation of 16.5 Gm. and a range of 15.0 to 18.6 Gm. They stated that a decrease in oxygen supply causes an increase in red blood cells, that the normal hemoglobin level of cord blood should be 14.8 Gm. with a red cell count of 4.0 million, and that the higher the level above 14.8 Gm. the greater the lack of oxygen.

A level at birth which should be considered as indicative of anemia has never been established. Allen and Diamond[4] state that in an infant with a positive Coombs test they consider a hemoglobin below 15.0 Gm. in capillary blood or 13.0 Gm. in venous blood an indication for immediate exchange transfusion. These are 4 to 5 Gm. below the figures generally considered normal and are probably as satisfactory as any.

During most of fetal life, the liver is the principal site of erythropoiesis. In the young embryo, mesenchyme in many areas produces both red and white cells, but this capacity is normally lost early in fetal life. Only relatively late in pregnancy, as the bones reach a considerable size, is the marrow sufficiently extensive to be responsible for the development of appreciable numbers of red blood cells. Even at term, small foci of erythropoiesis are still present in the liver; these may be more numerous in extremely large babies (over 4,500 grams) than in the average infant of 3,450 grams in weight.

An interesting phenomenon is the extreme rapidity with which erythropoiesis ceases in the liver after birth. This is true of even the small premature infant. No matter how low the birth weight, immature cells are usually absent after 3 or 4 days. This is also true when prenatal blood formation in the liver has been excessive as in severe erythroblastosis; even though the infant remains anemic and a need for large numbers of blood cells persists, the liver ceases its erythropoietic activity within a few days after birth. This can rarely be demonstrated today since treatment is generally instituted immediately after birth and infants seldom die, but we observed it frequently in the days before exchange transfusions. This was the principal reason why the diagnosis of erythroblastosis was often not made at autopsy in spite of severe jaundice; it was thought that erythropoiesis in the liver and other organs was a cardinal symptom of erythroblastosis and that it was necessary for the diagnosis.

The hemoglobin of the fetus differs from that of the adult in being more resistant to the denaturing effect of alkali or salicylate and less resistant to urea. The hemin is identical in the two groups so that the difference must be in the protein portions. It has been suggested that structurally identical hemin combines with the particular protein available at the site of chromoprotein fabrication. It combines with "liver" globin responsible for the properties of fetal hemoglobin and "red bone marrow" globin responsible for those of adult hemoglobin.[5]

Fetal-type hemoglobin comprises 70 to 80 per cent of the total hemoglobin at birth; it gradually diminishes until by the end of the third month it normally makes up less than 10 per cent of the total. In certain anemias, particularly thalassemia and sickle-cell anemia, higher proportions of fetal hemoglobin persist throughout life.

If fetal hemoglobin indicates formation in the liver, it is surprising that in erythroblastosis where, in severe cases, erythropoietic activity in the liver is very pronounced, fetal hemoglobin is reported to be decreased. Schulman and Smith[33] found fetal hemoglobin in 33 erythroblastotic infants to be significantly lower than in 96 normal infants, and they concluded that among infants born with normal hemoglobin levels fetal capacity to synthesize adult hemoglobin in amounts greater than normal appears to be an important mechanism in preventing anemia.

ANEMIA FROM FAILURE OF BLOOD FORMATION

Anemia which is demonstrable at birth and which is a direct result of failure of erythrocyte formation during intrauterine life is rare; the great majority of cases of anemia are the result of blood loss or of a failure to compensate for blood destruction. Although hypoplastic anemia without malformations has been described, Fanconi's syndrome is more common, and even here anemia is often not apparent until the child is several weeks or months of age. In Fanconi's syndrome[6, 7] malformations of various parts of the body, especially of the bones, are generally present and the condition is thought to be a defect of mesodermal tissues. The anemia is of the hypoplastic type and the bone marrow reveals marked hypocellularity. Formation of leukocytes and platelets also is often depressed. The condition is often familial and the reported cases suggest a recessive form of inheritance.*

ANEMIA FROM LOSS OF BLOOD

Vasa Praevia.—The most clear-cut and best-documented loss of blood before birth is from rupture of a velamentous vessel present in fetal membranes overlying the cervix. In a velamentous insertion of the umbilical cord, one or more vessels course through the membranes for varying distances between the placenta and main trunk of the umbilical cord. Even though they are more susceptible to damage than when embedded in Wharton's jelly, they are, nevertheless, injured very infrequently. On rare occasions the cord may arise in the membranes opposite the placenta and a vessel may occasionally cross the dilated cervix and be stretched to the point of rupture by the descending head; more often the vessel slips aside and the infant is delivered uneventfully.

Very infrequently, in fact as far as the records show and in my recollection only once, has rupture of such a vessel led to death of the fetus from exsanguination in the past 25 years at the Chicago Lying-in Hospital, and only on very few occasions has rupture led to enough blood loss to be responsible for anemia.

Hemorrhage From Vessels on the Surface of the Placenta.—Occasionally the amniotic fluid is bloody when

*See also *Pure Red-Cell Anemia* in this issue.

membranes are ruptured or blood may be found, usually only in small amounts, between the amnion and chorion. Ordinarily, in such instances, small injuries can be identified in vessels on the surface of the placenta. They are rarely of sufficient extent to be responsible for anemia.

Hemorrhage From Villi.—The total extent of the vessels within the villi is enormous. Even in terminal villi, several vessels can be seen when the villus is viewed in cross section. They are made up of only a single layer of endothelial cells, are surrounded by a small amount of mesoderm, and the outer surface of the villus is covered during the latter two-thirds of pregnancy by a single layer of syncytial cells. It is assumed that these thin-walled vessels within the villi may break and fetal blood may escape into the maternal circulation, but this assumption is extremely difficult to prove.

In practically all placentas after the middle of pregnancy, small amounts of a homogeneous eosinophilic material are found on the surface of occasional villi. Sometimes this is on the surface of individual villi and sometimes several villi are embedded in a single mass, or occasionally the masses are large enough to be visible grossly and are designated infarcts. Such areas have been described as being caused by local hemorrhage from a villus, but that this is their origin has not been actually proved.

Areas resembling thrombi are present in a few placentas; these have been described as proof of fetal hemorrhage. However, in my experience such areas are always composed of maternal and never of fetal blood. These areas are located centrally within cotyledons, are roughly cuboidal, never contain villi, and rarely exceed 1 cm. in diameter. At one time I examined 1,600 placentas and found 56 such areas in which blood cells could still be typed. Maternal and infant blood were also typed.[8] In every one of the 12 cases in which mother and infant were of different blood groups, the cells of the thrombus were of the same group as the mother.

The demonstration of fetal blood cells in the maternal blood by means of differential agglutination would give the most positive evidence of passage of fetal cells from villi to maternal circulation. Because of the great difference in blood volumes, an extensive loss of blood would be necessary before fetal blood could be detected. A few cases have been reported in which significant amounts of fetal blood were thought to have been demonstrated in maternal blood, but more cases will be necessary before this can be regarded as irrefutable evidence.[9] More refined techniques are being developed which may make it possible to detect smaller amounts of foreign blood and to positively prove fetal bleeding into maternal circulation by the direct demonstration of fetal cells.

Indirect evidence of some degree of hemorrhage from fetal vessels is attributed to the fact that some women become immunized to the Rh factor when they themselves are Rh negative and the fetus is Rh positive. This must be viewed, however, in light of the fact that evidence of immunization almost never occurs in a first pregnancy. If leakage of fetal blood

during pregnancy is the cause of immunization, it must be assumed that (1) antibodies are formed too slowly or too late in a pregnancy to affect the fetus of that pregnancy, or (2) that fetuses of a first pregnancy differ in some unknown way from fetuses of other pregnancies so that they are incapable of being adversely affected by antibodies.

It seems to me that a much more probable cause of immunization is passage of fetal blood into the maternal circulation during labor or delivery and evidence of immunization is not proof that fetal blood cells enter the maternal circulation normally. When the placenta is torn and villi are disrupted during the placenta's separation from the uterine wall, it would seem that fetal blood must almost invariably enter the wide-open maternal sinuses present immediately beneath the placenta. Villi may sometimes be injured during spontaneous delivery of the placenta, but they are always severely traumatized when the placenta is manually removed either following delivery through the vagina or by cesarean section. In association with cesarean section, an additional hazard is encountered when the placenta is incised because of its position on the anterior wall of the uterus. Under such circumstances the fetus loses some blood, but apparently this is rarely of sufficient amount to be injurious.

Loss of Blood From One Twin to the Other.—In the monochorionic placentas of mono-ovular twins, anastomoses between blood vessels of the portions belonging to the two twins is of common occurrence. This ordinarily produces no disturbance and both twins maintain separate circulations. It seems, however, on rare occasions that a disproportionate amount of blood may accumulate in one circulation at the expense of the other. This has been described on rare occasions, and we have observed a pair of twins in which one twin at birth had a hemoglobin of 8.5 Gm. with an erythrocyte count of 2.04 million while the other had a hemoglobin of over 20 Gm. On the second day of life the hemoglobin of that twin was 25.8 Gm. and the erythrocyte count was 7.82 million. One portion of the placenta was extremely pale and contained little blood; the other was dark red purple and all villi were distended with blood.

ANEMIA FROM BLOOD DESTRUCTION

The two principal recognized causes of destruction of antenatal blood are infections and antibodies specific for the blood cells of the fetus. Evidence of blood destruction varies depending on the extent of destruction and probably on the ability of the fetus to compensate for blood loss. An increase in bilirubin in tissues and circulating blood and an increase in erythropoiesis constitute the two main findings substantiating such a diagnosis. While the fetus is still in the uterus, bilirubin may become elevated in the fetal blood but the greater part seems to be transported across the placenta to the maternal circulation. The level is practically never more than 8 to 10 mg. per cent in cord blood, although it may become 2 or 3 times as high as soon as the infant loses this means of elimination. The greater part is indirect bilirubin, indicating a hemolytic process. However, occasionally in a very severely

affected stillborn fetus or in an infant who dies almost immediately after birth, hepatic cells may contain large amounts of hemosiderin, bile capillaries may be distended by thrombi, and rarely there may even be an increase in intralobular connective tissue, indicating that even before birth an obstructive process might be going on in the liver.

When the red blood cells are destroyed at an abnormally rapid rate before birth, erythropoiesis in the liver is immediately increased. If the need becomes excessive and the liver cannot compensate, other potential sources of erythropoiesis are called into action. Maximow described pericytes (which are totipotential connective tissue cells) in the immediate vicinity of small blood vessels which are capable, under stress, of producing blood cells. Erythropoiesis first becomes evident in the kidneys, especially in the vicinity of blood vessels at the apices of the pyramids, and later may be found around the blood vessels in any organ or tissue.

The degree to which the spleen is involved is variable. Sometimes the kidneys and other organs are the site of marked erythropoiesis while the spleen shows only small numbers of immature red blood cells; on the other hand, both the pulp and the sinuses may be filled with extremely immature cells.

A reduction in lymphoid tissue usually accompanies excessive erythropoiesis. The thymus is decreased in size, owing especially to a reduction in cortical lymphocytes, lymph nodes are small, malpighian corpuscles disappear from the spleen, and lymphoid tissue in general is inconspicuous. On rare occasions Peyer's patches and solitary follicles in the intestine are replaced by areas of erythropoiesis.

The extent to which potential centers of erythropoiesis are stimulated seems to be directly related to the destruction of red blood cells. When the need for replacement is mild, only the liver and bone marrow become more active, the majority of cells are not released into the circulation until nuclei have been lost; the only evidence of increased activity is an increase in reticulocytes. As the need becomes greater and other potential areas of erythropoiesis become active, younger cells escape into the circulation and at times cells even undergo mitosis in the peripheral blood. The proportionate number of immature cells and the degree of immaturity of cells in the circulating blood can be directly correlated with the extent of erythropoiesis in body tissues. The findings on histologic examination of tissues can be foretold almost invariably by examination of the peripheral blood in the newborn infant. The barrier that holds back nucleated red blood cells in the liver and in the bone marrow seems to be missing in erythropoietic centers in other tissues, and cells in such areas have free access to the circulation. In fact, the examination of such foci of erythropoiesis makes one wonder whether they are of aid in producing oxygen-carrying cells or whether the cells escape immediately into the peripheral circulation, thereby increasing only the number of nucleated forms.

The infants who at birth have the lowest hemoglobin and erythrocyte counts have also the most immature forms in greatest number in the peripheral blood. These extreme changes

in the blood are generally accompanied by severe edema (fetal hydrops), and the infants practically never survive. Not only may the hemoglobin be as low as 3 to 5 Gm. and the blood filled with extremely immature forms but the total volume of circulating blood generally seems greatly reduced. On several occasions we have tried unsuccessfully to get sufficient blood for chemical determinations from the heart and peripheral vessels at autopsy; it seems as if all the fluid was in the tissues with little remaining in the blood vessels. It is in such infants, too, that evidence of antenatal biliary obstruction is most common.

This extreme demand for replacement of red blood cells produces a truly remarkable picture in smears of peripheral blood. Nucleated cells in all stages of development are present. Generally there are many large basophilic erythroblasts and a scattering of stem cells with finely granular chromatin and multiple nucleoli and occasionally with mitotic figures. Non-nucleated cells are extremely variable in size, with macrocytes and microcytes both present in large numbers. Phagocytes containing red blood cells are occasionally seen.

Evidence of abnormal erythropoiesis can rarely be found before the fetus reaches 28 weeks' gestation, and it seems probable that blood destruction prior to this time is not great enough to stimulate increased blood formation. Consequently, it is assumed that most fetuses are not affected by maternal antibodies until fairly late in pregnancy; but in some instances, at least, there is contrary evidence in the placenta. In cases of severe erythroblastosis, the cytotrophoblast covering the surface of the villi persists until term, whereas normally it is lost before the end of the first trimester of pregnancy. When the destruction of blood cells is mild, the placenta may appear normal, but when it is severe, the placenta is always enlarged, the villi are bulbous and edematous, and the vessels within individual villi are often increased in number and are often almost completely filled with immature red blood cells.

When blood destruction is severe, plasma proteins are usually decreased, and in severely hydropic infants, they may be as low as 2 to 2.5 mg./100 ml. It has been suggested that heart failure as a result of severe anemia might be the cause of the edema; although this may contribute, more often the primary cause appears to be hypoproteinemia.

The principal cause of blood destruction is a result of an antigen-antibody reaction between the Rh positive cells of the fetus and antibodies derived from the maternal blood. Other similar reactions may result from interaction of other varieties of blood cell antigens and specific antibodies. These include Rh subgroups, A, B, Kell, Duffy, and so on.

Less common causes of fetal blood destruction are syphilis and the salivary gland virus. Both may be responsible for clinical and pathological changes identical to those resulting from a direct antigen-antibody reaction.

ANEMIA FROM MATERNAL IRON-DEFICIENCY ANEMIA

In normal infants hemoglobin production for the first several months utilizes iron obtained transplacentally, and not until 3 to 4 months of age

can any dietary iron be identified in blood cells.[10] If the mother has an iron-deficiency anemia during pregnancy, the red cells of the infant are normal at birth but because of inadequate stores of iron, iron-deficiency anemia generally appears within a few months of birth.*

ANEMIA FROM GENETIC CAUSES

A large group of anemias, many of them familial and some with well-established hereditary patterns, have been identified. Although congenital in nature, few give any antenatal evidence of their existence, although, occasionally, almost any may give symptoms in the first 24 hours of life. However, most often they do not manifest anemia until later infancy or childhood.

Thalassemia (Cooley's Anemia).—This is a hypochromic anemia with target cells, decreased osmotic fragility, and a survival time of erythrocytes as low as 18 to 20 days. In one child observed at 4 months of age, the hemoglobin was 4.6 Gm./100 ml. and the red blood count was 2.31 million. The disease appears to become manifest only when the thalassemia trait is in a homozygous form. When one gene for thalassemia and one for sickle-cell anemia are present, a disease indistinguishable from sickle-cell anemia is produced.[11] As much as 90 per cent of hemoglobin may be of the fetal type and homozygous thalassemia, and it has been suggested that the anemia may be due to an inability to change over from the fetal to the adult mechanism of synthesis.

Hemoglobinopathies.—Abnormal hemoglobin syndromes show an association of some one of the abnormal hemoglobins, S, C, D, or E, in combination with varying amounts of F (fetal) and A (adult) hemoglobins.[20] The S hemoglobin is found in the sickle-cell trait, in pure sickle-cell anemia, and in sickle-cell anemia combined with thalassemia, hemoglobin C, or hemoglobin D disease. It seems that all abnormal hemoglobins may be found as a "trait" when heterozygous and as a "disease" when homozygous. When heterozygous, any two may be combined. Various genetic combinations may produce hemoglobin C trait, homozygous hemoglobin C disease, sickle-cell hemoglobin C, thalassemia hemoglobin C, hemoglobin D trait, sickle-cell hemoglobin D, and so on. Reported, but less well substantiated, are other abnormal hemoglobins designated G and H.[21]

Hereditary Elliptocytosis.—This anemia is diagnosed by finding a high percentage of elongated cells in stained smears and wet preparations of blood from the patient and from other members of the family, including one parent.[12] In many individuals it appears to be a benign anomaly with no associated symptoms, although erythrocytes may have a shortened life span which is responsible for hemolytic anemia. Severe anemia has been reported in infants as early as 2 months of age.

Congenital Spherocytosis (Familial Acholuric Jaundice, Congenital Hemolytic Anemia).—This is a hemolytic anemia occurring in both Negroes and Caucasians, characterized by unusually globular microcytes and an increased osmotic fragility as a result of a dominant hereditary defect. When symptoms are present at birth, causative factors must have been operating in utero. According to

*See also *Considerations of Iron Metabolism in Early Infancy* in this issue.

Shapiro and colleagues,[13] hemolysis, anemia, and jaundice are not present at birth but may appear within the first 24 hours. Gellis[14] has reported observing 2 infants with early jaundice who required exchange transfusions.

Congenital Nonspherocytic Hemolytic Anemia.—This is an intrinsic corpuscular defect as in thalassemia and spherocytic anemia, although the nature of the abnormality is unknown; as in the others, extracorpuscular facors may contribute to the total hemolytic process. It has been suggested that a defect of liver metabolism may be present, resulting in macrocytosis, perhaps due to abnormal porphyrin metabolism. Jaundice within 24 hours, an enlarged liver, and hemoglobin of 12.2 Gm. at birth, as described by Krivit and coworkers,[15] may be confused with erythroblastosis. Bruton, Crosby, and Motulsky[16] reported an infant with 6.9 Gm. hemoglobin, 77,500 nucleated red blood cells per c.mm., and a bilirubin of 9.8 mg./100 ml. whose hemolysis persisted in spite of an exchange transfusion and who finally was benefited by splenectomy. Hepatosplenomegaly and jaundice are present. The red cells are normochromic, normocytic, and occasionally macrocytic. Basophilic stippling may be noted. Anemia becomes progressive and kernicterus may be associated with it. The patterns of inheritance of this form of the disease are believed to be mendelian dominant.

Atypical Congenital Hemolytic Anemia.—Certain of these anemias as reported by Brafield and Reef[17] may present early jaundice. They report this condition in siblings, one of whom had enlarged liver and spleen, anemia, jaundice, and bilirubin of 10 mg./100 ml. 4½ hours after birth, the other was first seen at 10 weeks of age and had a hemoglobin of 4.8 Gm. Splenectomies were performed in both with beneficial results as far as obviating the need for further blood transfusions, although the hemolytic process was not halted.

Familial Nonhemolytic Jaundice With Kernicterus.—This entity, first described by Crigler and Najjar[18] in 1952, is diagnosed by the appearance of jaundice on the first or second day. The diagnosis is based on a pronounced elevation of indirect serum bilirubin and delayed excretion of injected bilirubin. In most infants the jaundice persists, and bilirubin may rise to 40 mg./100 ml. or more. Red cells, osmotic fragility, reticulocyte counts, and urobilin excretion are normal. Liver biopsies may show some accumulation of bilirubin in a few bile canaliculi and a little periportal fibrosis. The disturbance is thought probably to lie in the pathophysiology of hyperbilirubinemia dissociated from extraneous factors such as antibodies, anoxia, or hemolysis. Inheritance appears to be as a mendelian recessive. A milder form of kernicterus is described as mendelian dominant.[19]

Congenital Methemoglobinemia.—This was first observed in 1844, and according to Dine[22] over 100 cases have now been reported. It is an inborn error of metabolism that may or may not be familial, and when familial, it has been described both as a dominant and a recessive state. In this condition 20 to 40 per cent of hemoglobin is in the methemoglobin

form instead of the normal 0.01 to 0.08 per cent and is responsible for a lavender-blue cyanosis which is generally present from birth. A decrease in cyanosis following administration of reducing substances such as ascorbic acid or methylene blue differentiates this condition from congenital heart disease. It can be definitely identified by finding a typical spectral band at 630 γ which disappears after the addition of potassium cyanide. (This differentiates it from sulfhemoglobin which has a spectral band at 618 γ that remains fixed after addition of potassium cyanide.)

POLYCYTHEMIA

An increase in the number of erythrocytes in the circulation at birth has not been described as a primary condition. Walker and Turnbull[3] have suggested that the concentrations ordinarily considered normal at birth may actually be higher than normal because of stimulation of erythropoiesis afforded by a presumed decreasing availability of oxygen as the fetus becomes more mature. The only infant we have observed with an erythrocyte count appreciably higher than that ordinarily considered normal was one of twins mentioned earlier, where blood from one twin presumably drained into the placental circulation of the other.

LEUKEMIA

Congenital leukemia is a rare but well-recognized form of this disease which, according to Pierce,* has been reported only 45 times. We have observed it only once in 27 years in the Chicago Lying-in Hospital. This infant has been previously described as presenting a characteristic clinical and histologic appearance. At birth, multiple flat and elevated nodules were present in the skin, the spleen and liver were markedly enlarged, and the circulating blood contained innumerable immature white blood cells that were interpreted as myelocytes. All organs were heavily infiltrated with similar cells and they were responsible for the tumefactions in the skin. To some extent, the cells were in the same locations occupied by erythroblasts in severe erythroblastosis and probably arose from the same cells that under other circumstances differentiate into erythrocytes.

The principal differences in the location of immature cells in erythroblastosis and leukemia lie in the excessive hematopoiesis in the portal areas of the liver and the severe involvement of the dermis in leukemia. During early fetal life, zones of myelopoiesis surround the blood vessels and bile ducts in the portal areas. These gradually decrease in size and prominence, and at term only a few myeloid cells are generally present. In erythroblastosis, erythropoiesis is much more prominent in the vicinity of the sinusoids and the portal areas are not appreciably stimulated. Small foci of erythropoiesis may be present in the dermis in erythroblastosis, but to my knowledge infiltrates producing grossly visible lesions have never been described.

DISTURBANCES IN BLOOD COAGULATION

Hemorrhagic Disease of the Newborn Infant.—Controversy about this condition has raged for many years. The term in the past has been applied loosely to any infant exhibiting

*See *Congenital Leukemia* by Mila Pierce in this issue.

hemorrhage at any site within the first few days of life. Hematemesis and melena were the most commonly described symptoms, but bleeding from the stump of the umbilical cord into the lungs and into the intracranial cavity have often been included. It seems probable that many of these cases would have been more properly ascribed to cutting the cord too short and improperly ligating it, to trauma sustained during birth, to infection, and to fibrinopenia, thrombocytopenia, and so on.

It now seems well established, however, that in some cases of so-called hemorrhagic disease the symptoms result from a prolonged prothrombin time associated with a deficieney of vitamin K. Prothrombin time of all infants at the time of birth, regardless of weight, is lower than the normal adult and appears to be especially low in infants who have suffered from anexia. Prothrombin originates in the liver under the influence of vitamin K and occurs in the plasma among the globulins. The role of vitamin K seems to be primarily to maintain a normal level of prothrombin in the blood. Thordarson[34] reported that blood prothrombin concentration in pregnant women begins to rise from the third month of pregnancy, and by the time of normal delivery, maternal prothrombin levels are higher than in normal nonpregnant adults. Dam and his associates[23] found that especially low birth values were present when the mother had severe albuminuria, when she had received large doses of a barbiturate, or when there were pathologic changes in the placenta.

The factors governing the characteristic prothrombin level at birth are not known. Ordinarily the infant appears not to lack vitamin K at birth, but a physiologic hypovitaminosis sets in a few hours after birth. Dam and co-workers[23] have suggested that the fetus may not need vitamin K, possibly because prothrombin is formed in the liver. Vitamin K administered to the mother prenatally, according to Dam and his colleagues, working in Copenhagen, is not associated with an elevated level of prothrombin at birth but does prevent a postnatal fall. They found that following prophylatic administration of vitamin K to 10,876 mothers, the frequency of hemorrhages of all types was 274 per 10,000, while it was 363 per 10,000 among 22,371 infants of mothers who were not given vitamin K. Even the figure for infants of treated mothers is far higher than that seen in American cities for infants of nontreated mothers. This suggests that factors other than prothrombin must be important in production of hemorrhage in the newborn infant.

Congenital Absence of Fibrin.—This finding was discussed by Grossman and Carter[24] in 1957, in conjunction with a case report of the only infant delivered at the Chicago Lying-in Hospital on whom such a diagnosis has been made immediately after birth. They were able to find only 31 such cases reported in the literature. The condition is charactertized by uncoagulability of the blood from birth, but in spite of this, hemorrhagic manifestations are not usually severe. The reason for the lack of fibrinogen is not known. In a few cases siblings have been reported to have bleeding disorders, but rarely have they been proved to have afibrinogenemia. Typical laboratory findings are an indefinitely prolonged clotting and

prothrombin time with normal bleeding time, normal platelet count, and absence of clot retraction.

Thrombocytopenic Purpura.—This occurs in the newborn infant both as a spontaneous disturbance and as one secondary to disease in the mother.[25] When the condition is present in the mother, the infant's platelet count is low, generally because of transplacentally transmitted platelet antiagglutinins. Schoen and co-workers[26] reported a case in which both the mother and the newborn infant had platelet antiagglutinins and isoagglutinins for each other's platelets. When the mother is not thrombocytopenic, the cause is generally thought to lie in isoimmunization of the mother to the infant's platelets, although a congenital decrease in megakaryocytes has also been suggested as a possible cause. As soon as the exogenously formed antiplatelet factor disappears, symptoms of purpura cease. No histologic abnormalities are detectable in any variety.

Thrombocytopenia also occurs in the newborn infant in conjunction with some hemolytic anemias, especially erythroblastosis and giant hemangiomas.[27, 28]

The occurrence of quinine-induced thrombocytopenic purpura in a mother and a newborn infant has been reported by Mauer and associates[29] and it is suggested that the condition in the infant resulted from the transplacental passage of antibody and quinine from the mother. They believe the possibility of drug sensitization should always be investigated when neonatal thrombocytopenia cannot be explained by other means.

Hemophilia.—Hemophilia is a hereditary hemorrhagic disease characterized by a delay in coagulation of blood which is responsible for a tendency to bleed. Platelet count, clot retraction, fibrinogen and prothrombin concentrations, tourniquet test, and bleeding time are normal. The clinical picture can be produced by a deficiency of at least 3 clotting factors: (1) antihemophilic globulin (AHG), (2) plasma thromboplastin component (PTC or Christmas' factor), or (3) plasma thromboplastin antecedent (PTA). Both AHG and PTC are inherited as sex-linked recessive mendelian traits transmitted by females and affecting males. PTA is not sex linked and may affect either males or females. The bleeding tendency may be present from birth and may be first detected by bleeding from the cord, after circumcision, or after slight injury.[30] In the more severe form, the danger of fatal hemorrhage in infancy is very great.

GLOBULIN DEFICIENCIES

Agammaglobulinemia.—It may be found in 3 forms according to Gitlin and his colleagues[31]: (1) a physiologic or transient form occurring in infants, (2) a sex-linked recessive character occurring only in males, and (3) an acquired form occurring in adolescents or adults. The congenital and acquired forms are due to an absence of plasma cells with a consequent deficiency of gamma globulin and failure of antibody synthesis. This failure appears to be permanent. These investigators have found that failure of synthesis of gamma globulin was not complete and that other plasma proteins are generally also either absent or markedly deficient. The clinical syndrome consists of extreme susceptibility to infection, low

to absent circulating gamma globulins and antibodies, immunologic unresponsiveness to antigenic challenge, and clinical improvement with replacement gamma globulin therapy.[32]

SUMMARY

Blood diseases with clinical manifestations at birth consist largely of anemias resulting from blood loss by hemorrhage or by cell destruction. Hereditary diseases, although determined at conception, only rarely produce symptoms at birth.

Pathologic changes in tissues that occur during intrauterine life are largely limited to the abnormal stimulation of erythropoiesis which results from severe blood destruction. The most common causes are maternal isoimmunization to Rh and the salivary gland virus. Congenital leukemia is rare and is characterized by massive myelopoiesis in skin, connective tissue, and all organs.

REFERENCES

1. Potter, E. L., and Bernstein, H. E.: Hematologic Studies on Children of the Rh-negative Women Compared With Those of Rh-positive Women, J. PEDIAT. **32**: 246, 1948.
2. Smith, C. A.: The Physiology of the Newborn Infant, ed. 2, Springfield, 1951, Charles C Thomas, p. 118.
3. Walker, J., and Turnbull, E. P. N.: Hemoglobin and Red Cells in the Human Fetus and Their Relation to Oxygen Content of Blood Vessels of the Umbilical Cord, Lancet **2**: 315, 1953.
4. Allen, F., and Diamond, L. K.: Erythroblastosis Fetalis, Boston, 1958, Little, Brown & Co.
5. Gardikas, C., Scott, D. G., and Wilkinson, J. F.: Observations on Fetal Hemoglobin, Arch. Dis. Childhood **28**: 38, 1953.
6. Pearson, H. A., and Cone, T. E.: Congenital Hypoplastic Anemia, Pediatrics **19**: 192, 1957.
7. Burgert, E. O., Kennedy, K. L. J., and Pease, G. L.: Congenital Hypoplastic Anemia, Pediatrics **13**: 218, 1954.
8. Potter, E. L.: Intervillous Thrombi in the Placenta and Their Possible Relation to Erythroblastosis Fetalis, Am. J. Obst. & Gynec. **56**: 959, 1949.
9. O'Connor, W. J., Shields, G., Kohl, S., and Sussman, M.: The Occurrence of Anemia of the Newborn in Association With the Appearance of Fetal Hemoglobin in the Maternal Circulation, Am. J. Obst. & Gynec. **73**: 768, 1957.
10. Smith, C. A., Cherry, R. B., Maletskos, C. J., Gibson, J. G., Roby, C. C., Caton, W. L., and Reid, D. E.: Persistence and Utilization of Maternal Iron for Blood Formation During Infancy, J. Clin. Invest. **34**: 1391, 1955.
11. Banks, L. O., Scott, R. B., and Simmons, J.: Studies in Sickle Cell Anemia: Inheritance Factor, Including Effect of Interaction of Genes for Sicklemia and Thalassemia, A. M. A. Am. J. Dis. Child. **84**: 601, 1952.
12. Josephs, H. W., and Avery, M. E.: Hereditary Elliptocytosis Associated With Increased Hemolysis, Pediatrics **16**: 741, 1955.
13. Shapiro, C. M., Josephson, A. M., Rosengvaig, S., and Kauffman, A.: Hereditary Spherocytosis in the Neonatal Period: Diagnosis, Incidence, and Treatment, J. PEDIAT. **50**: 308, 1957.
14. Gellis, S.: Editorial Comment, in Year Book of Pediatrics, 1957 to 1958, Chicago, Year Book Publishing Co., p. 288.
15. Krivit, W., Smith, R. T., Marvin, J. F., Read, R., and Good, R. A.: Congenital Nonspherocytic Hemolytic Anemia: Two Non-familial Cases With Red Cell Survival Studies, J. PEDIAT. **49**: 245, 1956.
16. Bruton, O. C., Crosby, W. H., and Motulsky, A. G.: Hereditary Nonspherocytic Hemolytic Anemia Presenting as Hemolytic Disease of Newborn Infant, Pediatrics **13**: 41, 1954.
17. Brafield, A. J., and Reef, I.: Atypical Congenital Hemolytic Anemia, Arch. Dis. Childhood **32**: 93, 1957.
18. Crigler, J. F., and Najjar, V. A.: Congenital Familial Nonhemolytic Jaundice With Kernicterus, Pediatrics **10**: 169, 1952.
19. Childs, B., and Najjar, V. A.: Familial Nonhemolytic Jaundice With Kernicterus: Report of Two Cases Without Brain Damage, Pediatrics **18**: 369, 1956.
20. Alway, R. H.: Recognition of Abnormal Hemoglobin Syndromes, Lancet **75**: 215, 1955.
21. Zuelzer, W. W., Neel, J. V., and Robinson, A. R.: Abnormal Hemoglobins, Progress Hemat. **1**: 91, 1956.
22. Dine, M. S.: Congenital Methemoglobinemia in the Newborn Period, A. M. A. J. Dis. Child. **92**: 15, 1956.
23. Dam, H., Dyggvie, H., Larsen, H., and Plum, P.: The Relation of Vitamin K Deficiency to Hemorrhagic Disease of the Newborn, Advances Pediat. **5**: 129, 1952.

24. Grossman, B. J., and Carter, R. E.: Congenital Afibrinogenemia: A Report on a Newborn Infant Without Fibrinogen, J. PEDIAT. **50:** 708, 1957.
25. Harrington, W. J., Minnick, V., and Arimura, G.: The Autoimmune Thrombocytopenias, Progress Hemat. **1:** 166, 1956.
26. Schoen, E. J., King, A. L., and Duane, K. T.: Neonatal Thrombocytopenic Purpura, Pediatrics **17:** 72, 1956.
27. Good, T. A., Carnazzo, S. F., and Good, R. A.: Thrombocytopenia and Giant Hemangioma in Infants, A. M. A. Am. J. Dis. Child. **90:** 260, 1955.
28. Meeks, E. A., Jay, J. B., and Heaton, L. D.: Thrombocytopenic Purpura Occurring With Large Hemangioma, A. M. A. Am. J. Dis. Child. **90:** 349, 1955.
29. Mauer, A. M., DeVaux, W., and Lohey, M. E.: Neonatal and Maternal Thrombocytopenic Purpura Due to Quinine, Pediatrics **19:** 84, 1957.
30. Kay, W. R., and Kupfer, H. G.: Spontaneous Hemothorax in Hemophilia: Case Report and Discussion of the Hemophilia Syndromes, With Remarks on the Management of Hemothorax, Ann. Int. Med. **47:** 152, 1957.
31. Gitlin, D., Hitzig, W. H., and Janeway, C. A.: Multiple Serum Protein Deficiencies in Congenital and Acquired Agammaglobulinemia, J. Clin. Invest. **35:** 1199, 1956.
32. Martin, C. M., Bronstein, E., and Dray, S.: Agammaglobulinemia: Clinical Staff Conference at the National Institutes of Health, Ann. Int. Med. **47:** 533, 1957.
33. Schulman, I., and Smith, C. H.: Fetal and Adult Hemoglobins in Hemolytic Disease of the Newborn, A. M. A. Am. J. Dis. Child. **87:** 167, 1954.
34. Thordarson, O.: Hyperprothrombinaemia During Pregnancy, Nature **145:** 305, 1940.

"Since all things, both argument and ocular demonstration, show that the blood passes through the lungs and heart by the action of the (auricles and) ventricles, and is sent for distribution to all parts of the body, where it makes its way into the veins and pores of the flesh, and then flows by the veins from the circumference on every side to the centre, from the lesser to the greater veins, and is by them finally discharged into the vena cava and right auricle of the heart, and this in such a quantity or in such a flux and reflux thither by the arteries, hither by the veins, as cannot possibly be supplied by the ingesta, and is much greater than can be required for mere purposes of nutrition; it is absolutely necessary to conclude that the blood in the animal body is impelled in a circle, and is in a state of ceaseless motion; that this is the act or function which the heart performs by means of its pulse; and that it is the sole and only end of the motion and contraction of the heart."

William Harvey, M.D., "An Anatomical Disquisition on the Motion of the Heart and Blood in Animals."

From Cardiac Classics, by Frederick A. Willius and Thomas E. Keys

HEMOLYTIC DISEASE IN THE NEONATAL PERIOD AND EARLY INFANCY

MARION E. ERLANDSON, M.D., AND MARGARET HILGARTNER, M.D.
NEW YORK, N. Y.

AS IN any age group, hemolytic anemias in the neonatal period may be of an hereditary or acquired nature (Table I). However, clinical manifestations of these anemias may be altered by factors peculiar to the newborn or young infant, and additional hazards are imposed by a status of immaturity. This is true whether the etiology of the hemolytic defect is genetically determined and permanent or extrinsic and temporary.

The existence of a congenital hemolytic syndrome may or may not be apparent at birth. In general, those anemias which are dependent upon the production of an abnormal type of hemoglobin are obscured or delayed in onset by the presence of large amounts of fetal hemoglobin.[1, 2] Those disorders which are not accompanied by the formation of an abnormal hemoglobin may be manifest at or shortly after birth.

Several of the hereditary hemolytic anemias are usually recognized by a particular morphology or osmotic fragility of the erythrocytes. In the normal newborn infant, the erythrocytes are macrocytic and display increased resistance to hemolysis in hypotonic solutions of sodium chloride.[3, 4] Therefore, the characteristic blood picture of such syndromes is not always found in the neonatal period even when in vivo hemolysis is present.[61, 62] Interpretation of blood findings by comparison with those in normal infants rather than those in adults may serve to increase diagnostic accuracy.

Substances capable of inducing hemolysis may be passively acquired by a transplacental route.[5, 6, 135] In this case the source of the antibody or toxin is removed at the moment of birth. Therefore, the resulting disease in the infant will be transitory and will disappear permanently as the responsible agent is eliminated from the infant's body. In the case of antibodies the rate of decrease of evidence of hemolysis parallels the rate of natural decay of the gamma globulins.[5, 7] Since capacity for antibody production is limited in the young infant, actively acquired hemolytic anemias with an autoimmune mechanism will be uncommon in this age group.[8, 9]

Many toxic hemolytic reactions are mediated by the glutathione system.[146] It has been suggested that inadequate amounts of reduced glutathione allow certain chemical substances to produce changes in intracellular hemoglobin which result in the formation of Heinz bodies and ultimate cellular destruction. Since erythrocytes of newborn

From the Department of Pediatrics of The New York Hospital-Cornell Medical Center.

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infants demonstrate instability of reduced glutathione, they may be sensitive to such toxins after even limited exposure.[15,52]

Two major consequences of any hemolytic process are anemia and the production of bilirubin. Destruction of 1 Gm. of hemoglobin is believed to yield 35 mg. of bilirubin.[10] A decrease of 1 Gm./100 ml. of blood in the hemoglobin concentration of a baby weighing 3 kilograms will be the source of 105 mg. of bilirubin (assuming the blood volume to be 100 ml. per kilogram). It is apparent that a degree of hemolysis insufficient to produce marked anemia may produce considerable amounts of bilirubin. Enzymes in the liver which are necessary for the conjugation of toxic indirect-diazo-reacting bilirubin to direct-reacting bilirubin-glucuronide are deficient in the first several days of life.[14-17] Therefore, an accumulation of the indirect-reacting pigment may occur to some degree in the presence of normal destruction and to a greater extent with increased destruction of erythrocytes.[18-23] The amount and duration of bilirubinemia will then vary not only according to the amount of hemolysis but also with the particular maturity of this enzyme system in each individual infant.

TABLE I. CLASSIFICATION OF HEMOLYTIC DISEASE*

I. HEREDITARY HEMOLYTIC SYNDROMES
- A. Normal adult hemoglobin present
  1. Congenital spherocytosis
  2. Congenital nonspherocytic hemolytic anemia
  3. Elliptocytosis
- B. Normal fetal hemoglobin in an abnormal situation plus adult hemoglobin
  1. Thalassemia
- C. Abnormal hemoglobin syndromes
  1. Sickle cell anemia
  2. Other abnormal hemoglobins alone or in combination with S-hemoglobin or thalassemia

II. ACQUIRED HEMOLYTIC ANEMIAS
- A. Related to presence of antierythrocytic antibodies
  1. Passively acquired antibodies
     a. Erythroblastosis fetalis due to Rh, A, B, or other recognized antibodies
     b. Clinical picture of erythroblastosis fetalis without identification of known antibody or incompatibility
  2. Actively acquired autoimmune hemolytic disease
- B. Related to presence of toxins: Introduced either by transplacental route or by direct contact after birth
  1. Naphthalene-induced hemolytic anemia
  2. Hemolytic disease caused by water-soluble vitamin K analogues
  3. Anemia related to infection

*This modification of an etiologic classification of hemolytic anemias is applicable to diseases occurring in patients of all age groups. However, several diseases not ordinarily observed in the neonatal period and early infancy have not been included, i.e., malignancy, uremia, etc.

The incidence of kernicterus appears to be related to serum levels of bilirubin concentration and to the size of the infant, regardless of the etiology of the jaundice.[24-29] Additional evidence suggests an increased susceptibility of the central nervous system to toxic effects of indirect-reacting bilirubin during the neonatal period.[30, 31]

Infants demonstrating hemolytic defects either of a temporary or a permanent nature are subject to the dangers of hyperbilirubinemia in this early

period. Every infant with a hemolytic disease manifest at or shortly after birth requires consideration from the point of view of control of the level of this pigment. An exchange transfusion offers both removal of "destroyable" erythrocytes, i.e., removal of the source of the pigment, and also somewhat less efficient removal of bilirubin.[32, 33] Therefore, it is apparent that exchange transfusion may be indicated in the management of any hemolytic process occurring in the newborn period. In the majority of hemolytic diseases other than Rh erythroblastosis, anemia is not usually of great significance. The degree or rate of rise of bilirubinemia rather than anemia is the proper guide to the need for exchange transfusion.[6]

In the first several weeks of life, erythrocyte production is at a low level.[34] The exact stimulus to resumption of active erythropoiesis is unknown, but it has been suggested to be the attainment of a lowered hemoglobin concentration adequate to stimulate marrow production via levels of oxygen tension.[35] Prolonged hypoplasia of the bone marrow has been noted in infants with erythroblastosis.[36, 130] The mechanism of this prolongation is poorly understood. A number of infants in whom a hereditary hemolytic syndrome is present have been noted to have marked anemia between the ages of 1 and 3 or more months and yet stabilized with adequate, although subnormal, hemoglobin levels in later infancy and childhood (Fig. 1). A dubious capacity for marrow response in the presence of increased erythrocyte destruction and a great rate of increase in body size with its demand for an increase in blood volume may explain this pattern.

An understanding of the pathophysiology of each type of hemolytic disease in later childhood is mandatory to an appreciation of alterations in clinical manifestations in the newborn period and early infancy, and as a basis for decisions regarding management during this peculiar transitory phase of life.

HEREDITARY HEMOLYTIC SYNDROMES

Congenital Spherocytosis.—This disorder is marked by the genetically determined production of abnormal spherical erythrocytes.[37, 38] It is usually transmitted as a mendelian dominant characteristic.[39-41] However, in some instances a positive family history cannot be found.[41] This disease may occur in Negro as well as in white individuals.[42, 43]

The erythrocytes contain normal hemoglobin. They display an increased osmotic fragility when placed in hypotonic solutions of sodium chloride, and this fragility is further increased by incubation at 37° C.[38, 44, 45] It has been demonstrated that there is an abnormality of carbohydrate metabolism within the erythrocytes from patients with this disorder.[46, 47] These cells survive abnormally short periods of time in the circulation of any individual in whom the spleen is in situ.[38, 48] After removal of the spleen, erythrocyte survival is normal or only slightly impaired despite the continued presence of abnormal erythrocytes.[38, 44-46, 49, 50]

A great variability of clinical manifestations of this disorder exists in all age groups and among members of a single family.[41, 51] When a diagnosis

of congenital spherocytosis is made after the period of infancy, there is often a paucity of symptomatology resulting from the disease. Mild fatigability may be present or there may be no recognized symptoms. The value for hemoglobin concentration is often normal or only slightly lowered.

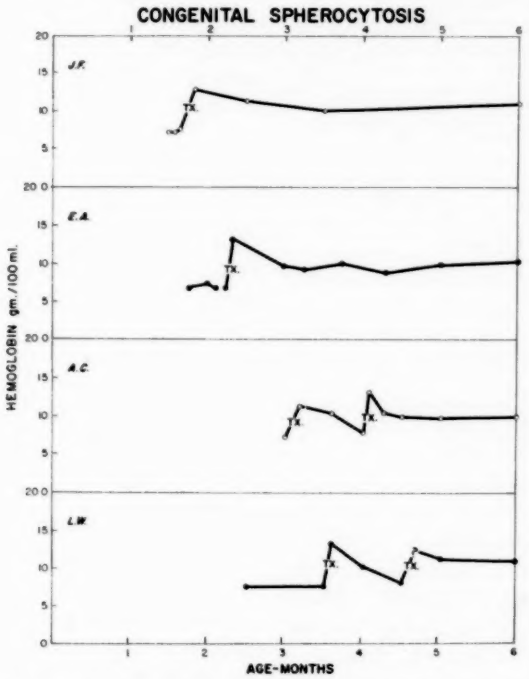

Fig. 1.—Four infants with congenital spherocytosis examined under the age of 3 months because of anemia. Each of these patients received one or two transfusions and subsequently maintained adequate levels of hemoglobin concentration without further transfusions and despite continued hemolysis.

More detailed studies reveal an abnormally small total erythrocyte volume (red cell mass) and a rapid rate of production of erythrocytes which has resulted in a partially compensated hemolytic anemia.[52] If erythropoiesis diminishes (hypoplastic or aplastic crisis), severe anemia will ensue.[53] Therefore, if such patients display few symptoms and if the parents are educated to observe for signs of increasing anemia, which may require supportive transfusion therapy, splenectomy may be safely deferred until later in childhood. Such management seems indicated in view of the increasing evidence that splenectomy in infancy and childhood imposes a risk of severe and overwhelming infection.*[54-59]

In addition to the previously mentioned variability of this disorder,

*It is suggested that in such patients splenectomy be performed between the ages of 7 and 9 years in order to minimize the risk of a major complication, gall bladder disease, the incidence of which increases past this age.[60] The need for earlier splenectomy may be indicated by the occurrence of persistent severe anemia or frequent crises.

several factors peculiar to early infancy may both influence the facility of diagnosis despite evidence of in vivo hemolysis and impose particular hazards during this period. These include the presence of macrocytic, fetal hemoglobin-containing erythrocytes which possess increased resistance to hemolysis in hypotonic sodium chloride solutions as well as the factors of liver immaturity, bone marrow hypoplasia, and increasing body size. Therefore, spherical erythrocytes may be difficult to recognize and osmotic fragility may be apparently normal, especially when interpretation is based on a comparison with the fragility of normal erythrocytes from adults.[58, 61, 62]

Congenital spherocytosis may be manifest immediately following birth[43, 57, 63-67] and has been recorded before the age of 6 months in 40 patients.[41, 43, 54, 57, 58, 61-69]

In the immediate neonatal period, anemia may occur but is usually not extreme and may be relieved by transfusion. However, accumulation of significant amounts of bilirubin may be dangerous. Neonatal jaundice has been reported by history or observation in 53 per cent of 43 infants with congenital spherocytic anemia.[58] Kernicterus has occurred as a result of this disease.[64, 79, 80] Although the hemolytic defect in the erythrocytes of such infants is permanent, the immaturity of the enzyme systems in the liver required for adequate conjugation of bilirubin is temporary.[17] Jaundice is not marked in these patients after the neonatal period. Therefore, exchange transfusion may be of great value in protecting the infant with spherocytosis from permanent damage occurring during a temporary hazardous period. Values for hemoglobin concentration do not constitute adequate criteria for this procedure.[6] The need for such therapy should be based on the level or rate of rise of serum bilirubin. In performing an exchange transfusion, a majority of the short-lived spherocytic cells are removed and replaced by erythrocytes with a normal life span. Only partial removal of bilirubin from the body is accomplished.[32, 33] Multiple exchange transfusions may be required to maintain adequate control of the bilirubinemia. Following this procedure and over a period of weeks, the normal donor erythrocytes will gradually be replaced by newly produced spherocytic cells. By the time this occurs, normal liver function will be present.*

A number of infants with congenital spherocytosis attract attention in the first several months of life because of pallor. Serial values for hemoglobin concentration in 4 such infants are presented in Fig. 1. In 3 infants, J. F., E. A., and L. W., the ages at the time of the initial observation were 6 to 10 weeks. One infant, A. C., was first seen at 3 months of age following an infection. Each of these infants was given 1 or 2 transfusions, following which adequate hemoglobin levels were maintained without further transfusion therapy. As may be noted by inspection of Fig. 2, such a clinical course of disease does not always ensue. In some infants severe anemia is persistent in the absence of repeated transfusion therapy. Of 5 infants

*In several instances a reduction of the value for serum bilirubin has been attributed to splenectomy performed in the neonatal period. It is difficult to ignore the fact that these operations were performed at an age when conjugation and excretion of bilirubin would be expected to increase by virtue of maturation of enzyme systems. The lessening of the icterus may have proceeded by virtue of this mechanism as well.[68]

with congenital spherocytosis under 3 months of age examined at The New York Hospital because of anemia, only one demonstrated a course of persistent anemia with related symptoms requiring multiple transfusion therapy. Of 5 infants recently described by Stamey and Diamond,[66] only one required early splenectomy by this criterion. Therefore, it would seem that many infants with this disorder demonstrate severe anemia in the first few months of life, but they can be supported with transfusion therapy through a limited period of infancy and subsequently attain a status in which anemia is minimal and symptoms are few. If the presence of an underlying hemolytic defect is recognized, splenectomy may be deferred under a regime of careful observation. Some infants will be persistently anemic and symptomatic and require a somewhat earlier operation. In the experience of the staff at The New York Hospital, the former is the usual and the latter the unusual pattern of disease.

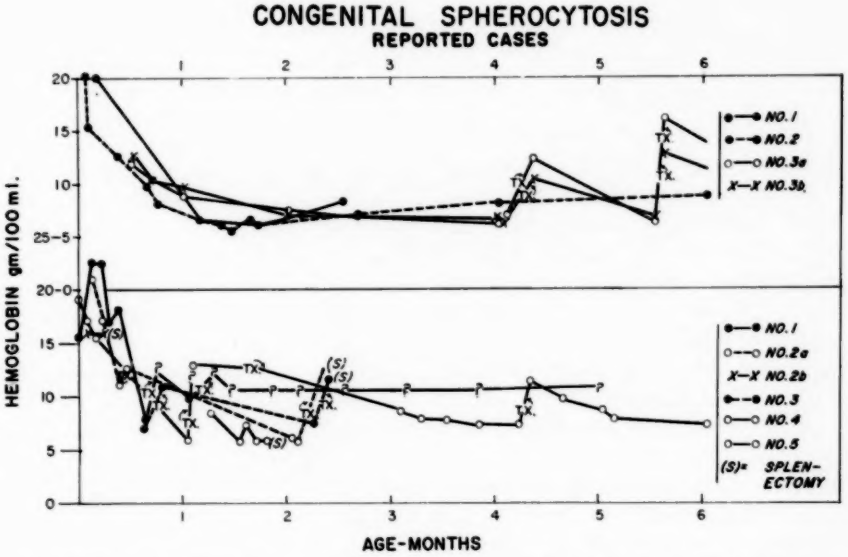

Fig. 2.—Reported cases of infants with congenital spherocytosis in whom serial values for hemoglobin concentration are available. ?, Later values in this infant were described as adequate, although numerical data were not recorded. References for these data are as follows. Upper chart: 1 = 68, 2 = 43, 3a and 3b = 58; lower chart: 1 = 63, 2a and 2b = 64, 3 = 62, 4 = 57, 5 = 76. Note accentuation of neonatal decline in hemoglobin concentration.

Congenital Nonspherocytic Hemolytic Anemia.—In 1950 Crosby[51] introduced this terminology to designate patients with a hemolytic anemia apparently similar in mode of genetic transmission and in hematologic and clinical expression to congenital spherocytic anemia but with several differences. Spherocytes are not found. Osmotic fragility of erythrocytes is normal. Splenectomy appears to be of little value.[81-88]

This disorder has been reported in the newborn period.[87] Clinical manifestations in infancy are similar to those described in congenital spherocytosis.[81, 82, 84-88] Principles of management are the same as in patients with congenital spherocytic anemia with the exception that splenectomy is not indicated for control of the basic hemolytic defect.

Elliptocytosis.—This hereditary anomaly of the erythrocytes is characterized by the presence of oval or elliptical cells in the blood smear. In many patients there is minimal or no evidence of hemolysis, and osmotic fragility of the erythrocytes is normal. In a few individuals microspherocytosis has been present in addition to the oval cells. In such instances an increased osmotic fragility of the erythrocytes may exist and a more severe hemolytic process may be apparent.[88] Elliptocytosis which was accompanied by jaundice in the neonatal period and a serious degree of anemia has been reported in a 1-month-old infant.[89] It is of great interest that both parents of this infant demonstrated elliptocytes in the peripheral blood smear. The findings in this family and that reported by Wyandt[90] suggest that a relatively asymptomatic condition may be representative of the heterozygous state and that homozygosity produces a severe hemolytic anemia. In the first patient evidence of hemolysis was minimal following splenectomy.

Thalassemia (Cooley's Anemia, Mediterranean Anemia, Erythroblastic Anemia.)—The gene for thalassemia behaves as an incomplete dominant characteristic.[91] In the heterozygous state findings consist of morphologic aberrations of the erythrocytes with or without mild anemia and minor splenomegaly.[92, 93] In the homozygous state abnormalities in the blood include: marked morphologic changes and hypochromia of the erythrocytes, normoblastosis, reticulocytosis, marked anemia, increased percentages of fetal hemoglobin, and increased amounts of iron in the serum with complete saturation of the iron-binding proteins.[94-96] The appearance of individuals with the homozygous disease is distinguished by the "Cooley's facies" which results from expansion of bone marrow spaces associated with increased erythropoiesis. A protuberant abdomen is related to massive enlargement of the spleen.[94] There is marked pallor. With progression of the disease and the use of transfusion therapy, diffuse pigmentation of the skin develops.[97, 98]

This is not an abnormal hemoglobin syndrome in the usual sense of the phrase. It is a disease in which a normal type of hemoglobin, i.e., fetal hemoglobin, occurs in an abnormal situation.[96, 99, 100] The principal physiologic defects are two: an abnormally rapid rate of destruction of erythrocytes and an inability to synthesize adequate amounts of normal adult hemoglobin.[101-103]

In most individuals in whom the heterozygous disease is present, the diagnosis is an incidental one made in later childhood or adult life. No disturbing symptoms are present.[92, 93] The majority of infants with the homozygous disease or thalassemia major (also known as severe Cooley's anemia) are presented for diagnosis well after the first 6 months of life when anemia has already resulted in pallor, interference with growth, and

symptoms of irritability and fatigability. At that time, all of the physical and blood findings referred to above are manifest.[94, 103] The presence of marked evidence of chronic disease in such patients has suggested that a much earlier diagnosis would have been possible and earlier transfusion therapy beneficial.

available in only one infant who subsequently developed the manifestations of severe Cooley's anemia. At 3 days of age, the value for hemoglobin concentration was 12.7 Gm./100 ml. of blood.[105] Although this value is abnormally low, it is not possible to state categorically that it represented an early manifestation of

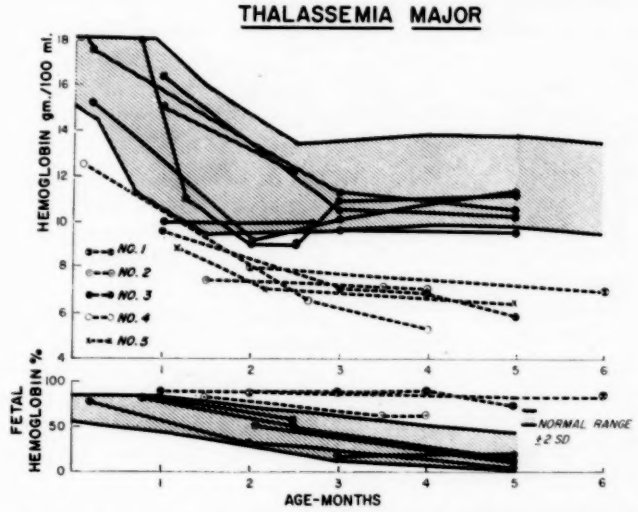

Fig. 3.—Progression of hematologic findings in 5 infants with homozygous Cooley's anemia or thalassemia major. Patients 1, 2, and 3 were observed at The New York Hospital. Patients 4 and 5 were reported by Banton.[105] Values for hemoglobin concentration, per cent of fetal hemoglobin, together with marked morphologic changes of the erythrocytes and normoblastosis in the blood smear permit a definitive diagnosis within the first 2 months of life. Normal ranges are from references 2 and 166. Solid lines represent normal or heterozygous infants having two heterozygous parents.

Production of the adult type of hemoglobin begins in or even before the third trimester of intrauterine life.[104] Although the major component of hemoglobin in the blood of normal newborn infants is of the fetal type, it is conceivable that some slight degree of anemia would be present in newborn infants homozygous for the thalassemia gene due to an inadequate production of adult hemoglobin.[1] Hematologic data soon after birth are

thalassemia major and not the result of some other etiologic mechanism.

Fawdry[106] is stated to have diagnosed severe Cooley's anemia in 3 infants by the age of 3 months. In two other infants reported in the literature, a similar diagnosis has been made at the same age.[105, 107] Fig. 3 demonstrates the development of severe Cooley's anemia in the neonatal period and early infancy. This figure represents a composite of the findings

in two infants described by Banton[105] and in three infants observed at The New York Hospital. It may be noted that values for hemoglobin concentration were already abnormally low in these infants when first observed between 1 and 2 months of age. (Immediate neonatal values are not available in 4 of the 5 infants.) The erythrocytes in the peripheral blood smear were morphologically abnormal in the infant examined at 3 days of age and in the 4 infants first examined within the first 2 months of life. The earliest morphological changes were similar to those noted in adults with the heterozygous disease. However, by 2 months of age, marked erythrocyte abnormalities were characteristic of the homozygous state and normoblasts had appeared in the peripheral blood. Splenomegaly was not present in the 1 infant examined at birth but became apparent and progressive in all 5 infants between 1 and 3 months of age. In the 3 infants in whom determinations of fetal hemoglobin were made, the values were above the normal range at ages of 1 or 2 months. From these data it is apparent that the presence of severe Cooley's anemia may be diagnosed within the first 2 months of life.

The need for transfusion therapy must be determined individually for each patient by the extent of the symptoms and signs resulting from the presence of this disease. Without the benefit of transfusion therapy, such infants have died by the age of 14 months.[108] R. M., in Fig. 3, demonstrated a Cooley's facies, irritability and marked pallor, and a hemoglobin concentration of 5.8 Gm./100 ml. of blood at 5 months of age. Transfusions were therefore required from that age. However, D. G., also in Fig. 3, displayed normal growth and development under careful observation until the age of 18 months, despite values for hemoglobin concentration of 6.5 to 7.0 Gm./100 ml. of blood. At that time she demonstrated irritability and interference with weight gain concomitant with a fall in value for hemoglobin concentration to 5.7 Gm./100 ml. of blood. Transfusion therapy was then initiated.

It would seem, therefore, that some manifestations of thalassemia major or severe Cooley's anemia may be present at birth. There is further progression of the disease during the period of early infancy to the extent that a diagnosis of thalassemia major is unequivocal between the ages of 1 and 2 months. The development of clinical manifestations may determine a desirability of transfusion therapy as early as 3 or 4 months of age in some patients, although the majority of infants with severe Cooley's anemia do not require supportive transfusions until some time after the first 6 months of life.

Sickle Cell Anemia.—This was the first disease in which the presence of an abnormal type of hemoglobin was recognized. Although the clinical syndrome accompanied by sickle-shaped red blood cells was recorded in 1910 by Herrick,[109] it was not until 1949 that the distinctive rate of electrophoretic mobility of sickle hemoglobin was demonstrated.[110] Subsequent extensive studies of other physical chemical properties of this hemoglobin have produced descriptions of tactoid formation and gelling phenomena which are intensified by reductions in oxygen tension or pH.[111, 112] A biochemical

aberration within the globin moiety has recently been documented.[113] These properties of S-hemoglobin are related to the sickling phenomenon observed in intact erythrocytes containing S-hemoglobin and vary with the amount of the abnormal hemoglobin present. this trait, sickle hemoglobin comprises 22 to 45 per cent of the total hemoglobin content, the remainder being of the normal adult type. No anemia results. In individuals homozygous for this condition, 76 to 100 per cent of the total hemoglobin is hemoglobin S,

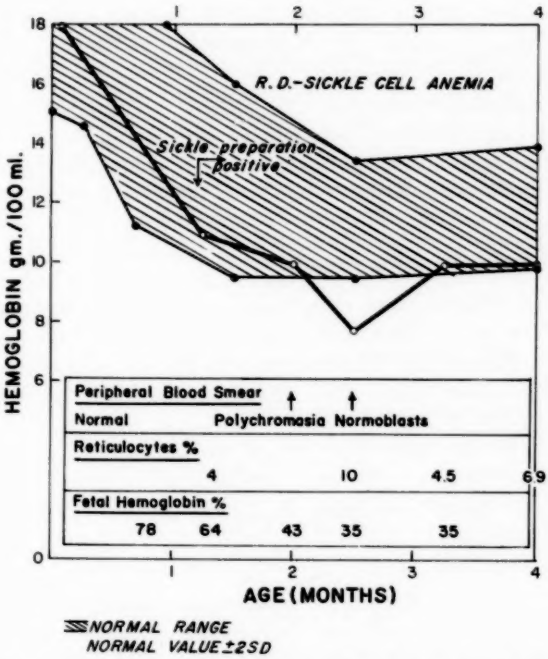

Fig. 4.—An infant observed from birth in whom sickle cell anemia developed. The sickle preparation was positive when first properly tested at the age of 1 month (see text regarding number of sickle cells in the newborn period). Reticulocytosis, polychromasia, and normoblastosis appeared during the first 2½ months of life. Subsequent values for hemoglobin concentration represent the status of a partially compensated hemolytic anemia. Normal hemoglobin range from reference 166.

Sickle cell disease is found primarily in Negroes but may occur in white individuals, particularly those of Mediterranean origin.[114, 115]

Identification and quantitation of the types of hemoglobin in erythrocytes from persons demonstrating the sickling phenomenon have served to further clarify the genetics of this disease. In individuals heterozygous for the remainder being of the fetal type.[116, 117] A hemolytic anemia results which is marked but is usually well tolerated. Although hemolysis is continuously present, the majority of patients require transfusions only infrequently and in the presence of an aplastic or hyperhemolytic crisis. The clinical picture of a chronic, partially compensated, hemolytic anemia is

punctuated by episodic painful "crises" which are thought to result from intracapillary sickling of erythrocytes.[118-120]

The incidence of the sickling phenomenon (sicklemia or sickle cell trait) is probably the same in the newborn Negro infants, 8 per cent, as in the Negro population at large.[120, 121] However, the number or per cent of sickle cells demonstrated in young infants is less than that demonstrated at later ages even in the same infant. It has been suggested that this is due to the effect of large amounts of fetal hemoglobins which are present in the younger age groups.[120, 121] In two infants who later were shown to have homozygous sickle cell disease, no anemia was present in the immediate neonatal period. Values for hemoglobin concentration were 17 Gm./100 ml. of blood at 4 days of age and 18 Gm./100 ml. of blood at 3 days of age, respectively[123] (Fig. 4).

On the basis of hematologic findings, seven patients described in the literature have been diagnosed as having sickle cell anemia between the ages of 1 and 3 months.[123-126] A similar diagnosis has been made in two such infants at The New York Hospital.*

Fig. 4 records the hematologic progression of the disease in one such infant. The sickle preparation was positive when first properly tested at the age of 1 month. The neonatal decline in level of hemoglobin did not appear unusually rapid but continued to a level of 7.6 Gm./100 ml. of blood at the age of 2½ months. Stabilization of the hemoglobin concentration then occurred at the value of 9.9 Gm./100 ml. of blood for 2 months and subsequently ranged between 9.8 and 7.6 Gm./100 ml. of blood. Reticulocytosis and normoblastosis became evident as the neonatal decline of hemoglobin concentration terminated, which suggested an increasing production of cells with a shortened life span and evidence of an erythropoietic response to a hemolytic process.*

A similar sequence of events has been described by Trowell and his colleagues[126] at a slightly earlier age. Anemia of 8 Gm./100 ml. of blood in the presence of normal erythrocyte morphology was first noted at 5½ weeks of age. This was followed in the next several weeks by the appearance of polychromasia and normoblastosis. Once the status of a partially compensated hemolytic anemia has been established in such infants, they must be considered susceptible to the chance occurrence of an aplastic type of crisis, during which there would be a marked increase in the degree of anemia.

Although the typical "painful crises" with bone, joint, or abdominal involvement have not been described in infants under 6 months of age, this does not necessarily indicate that such an episode cannot take place at this early age. Periods of irritability in these infants might be indicative of

*A definitive diagnosis of sickle cell anemia in early infancy is difficult by conventional paper electrophoretic methods. The large amounts of fetal hemoglobin make difficult the demonstration of small amounts of adult hemoglobin which would indicate the developing pattern of the trait rather than the homozygous disease. Shields and his associates[122] and MacIver and co-workers[127] have published results of paper electrophoresis of hemoglobin solution obtained by the use of 0.02 ionic strength veronal buffer (pH 8.6) rather than 0.06 ionic strength. Separation of hemoglobins A, F, and S appears more distinct when this buffer is used.

*Chromium[51] erythrocyte survival studies have been reported in two young infants with sickle cell anemia.[123] However, the presence of infection with its possible exaggeration of the hemolytic defect makes interpretation of the results difficult. Therefore, the degree of hemolysis occurring in young infants with sickle cell anemia has yet to be determined.

what would, at a later age, be recognized as a crisis. Although it is probable that residual fetal hemoglobin protects to some degree against intracapillary sickling, marked sickling of erythrocytes has been noted in the organs of young infants succumbing to intercurrent infections. Such findings during postmortem examinations have prompted the diagnosis of sickle cell disease in several patients at the age of 3 months.[126, 128]

It is of interest that McGavack and Nussbaum,[129] in a review of 173 patients with sickle cell anemia, found that symtoms were present by history in 50.8 per cent of patients before the age of 2 years. Trowell and associates[126] have pointed out the frequent occurrence of episodic osteopathy with swelling in the hands and feet in infants under 1 year of age with homozygous sickle cell anemia.

ACQUIRED HEMOLYTIC ANEMIAS

Those Related to the Presence of Antierythrocytic Antibodies.—The most frequent hemolytic anemia based on an immune mechanism is, of course, erythroblastosis fetalis. This disease is discussed elsewhere in this issue.[6] The offending antibodies are passively acquired. Since the source of the antibodies is removed by the process of birth, the hemolytic disease is of a limited duration which is determined by the initial intensity of the disease and by the rate of natural decay of the antibodies within the infant's body.[5, 7]

It should be mentioned that rarely hemolytic disease occurs in a newborn infant and follows the pattern of erythroblastosis, although the presence of known antibodies or a possible incompatibility between the mother and infant cannot be documented. Two siblings were observed at The New York Hospital in whom anemia, jaundice, spherocytosis, and reticulocytosis occurred in the neonatal period, with subsequent disappearance of all of these findings. The Coombs tests were negative, and recognized incompatibilities were not found. The presence of congenital spherocytosis was excluded in both parents and later in both children. There was no evidence of an acquired hemolytic anemia in the mother. This occurrence in 2 siblings between whom there was a year's difference in age, suggests the transmission of some substance, possibly an unidentified antibody, from the mother to these infants. This suspicion was strengthened by a positive reaction between trypsinated erythrocytes from the father and serum obtained from the mother following the delivery of the second affected infant.

Actively acquired autoimmune hemolytic anemias are rare in the first several months of life because of the infant's inability to produce antibodies readily.[7-9]

HEMOLYTIC DISEASE RELATED TO TOXINS

Naphthalene-Induced Hemolytic Anemia.—A severe hemolytic anemia in children following the ingestion of naphthalene was first described in the American literature by Zuelzer and Apt.[131] Anisocytosis, polychromasia, microspherocytes, fragmentation of erythrocytes, Heinz body formation, reticulocytosis, and normoblastosis were present in the peripheral blood. Although ingestion of naphthalene would not be expected to occur in earliest infancy, subsequent reports have demonstrated other modes of exposure. Two infants of ages 6 and 11

days, respectively, have acquired naphthalene intoxication following the use of diapers and clothes which had been stored in mothballs.[132, 133] In one infant the toxin was received via the placenta from the mother who had "smelled, sucked, and chewed" mothballs prior to delivery.[134, 135] Of twelve such infants and children reported, 8 have been Negro.[131-138]

The hemolytic process is actually produced by the metabolites of naphthalene which are alpha- and beta-naphthol and alpha- and beta-naphthoquinone.[136] It had previously been noted when beta-naphthol was used in the treatment of parasitic diseases that a hemolytic anemia appeared in some patients and that the incidence of this reaction suggested a racial and individual susceptibility.[139] A similar toxic hemolytic anemia has occurred in a limited portion of a Negro population receiving primaquine (an 8-aminoquinoline).[140] The primaquine-induced anemia is one manifestation of a multiple drug sensitivity.[141] Investigations of the mechanisms involved have produced insight into many drug reactions. The defect is localized within the erythrocytes of the susceptible individual, and Heinz body formation may be seen on incubation of the cells with the drug.[142, 143] The defect is most marked in the older erythrocytes which results in the production of a self-limited type of hemolytic process.[144, 145] The defect may be simulated in normal cells by the addition of a sulfhydril inhibitor.[146] Low levels of glutathione are found in erythrocytes of sensitive individuals, and this level may be further lowered by exposure to primaquine or acetylphenylhydrazine. This phenomenon is the basis of the so-called glutathione stability test.[147]

Reduced glutathione is apparently necessary to protect intracellular hemoglobin from the toxic effects of a number of drugs. Glutathione is reduced by an enzyme, glutathione reductase, in the presence of reduced triphosphopyridine nucleotide (TPNH). In individuals who demonstrate a genetically determined drug sensitivity, the basic biochemical defect is a deficiency of glucose-6-phosphate dehydrogenase, which is necessary for production of adequate amounts of TPNH.[148] A deficiency of glucose-6-phosphate dehydrogenase and instability of glutathione have also been demonstrated in individuals in whom naphthalene-induced hemolytic anemia has occurred.[134, 135]

Anemia with Heinz body formation due to naphthalene, instability of glutathione, and deficiency of glucose-6-phosphate dehydrogenase have been noted in some Caucasians, but the incidence is less than in Negroes where it occurs in 15 per cent of males and 2 per cent of females.[132, 138, 149-153] Genetic transmission is as an incomplete dominant characteristic with sex linkage.[149]

Instability of glutathione by some unknown mechanism (not by deficiency of glucose-6-phosphate dehydrogenase) is present in normal newborn infants as a transitory phenomenon.[134, 152, 153] Therefore, even "nonsensitive" newborn infants may be susceptible for a limited period of time to the number of drugs which are effective via this mechanism. Such drugs include naphthalene, acetanilid, sulfanilamide, thiosulfone (Promizole), phenylhydrazine, sulfoxone (Diasone), phenacetin, and the water-soluble vitamin K analogues[134, 141] (see below). These drugs in high concentrations may also have a toxic effect on normal erythrocytes.

Whether the biochemical defect is the transitory glutathione instability noted in normal newborn infants or a genetically determined enzyme deficiency, the hemolytic process, once induced, is temporary. In normal infants the instability of reduced glutathione is not apparent long after 95 hours of age.[153] In the sensitive individual, the defect is present in the older erythrocytes. Therefore, as these are destroyed and replaced by newly produced cells, hemolysis ceases. However, should the period of active hemolysis occur in the neonatal period, jaundice as well as anemia again warrants attention. Exchange transfusion or simple supportive transfusions may be necessary.

Water-Soluble Analogues of Vitamin K.—Because of their chemical structures as derivatives of naphthoquinone, several of these drugs were investigated in *adult* rats as early as 1940.[154, 155] At that time, adverse effects on hemoglobin concentration and erythrocyte count were noted when massive doses were employed. It was concluded that these drugs were not toxic when used in the usual therapeutic dosages. However, in 1953, Gasser[156] described a hemolytic anemia occurring in premature infants and characterized by the formation of Heinz bodies within the erythrocytes. These infants had received Synkayvite in amounts of 5 to 10 mg. daily. Subsequently, several authors suggested a relationship between an increase in the incidence of hemolytic anemia and kernicterus in premature infants and an increased routine dosage of this water-soluble analogue of vitamin K.[157, 158] Observations of bilirubin levels have demonstrated that there is also a correlation between the concentration of bilirubin in the serum and the administration of the larger dosages of this preparation in newborn infants.[159-161] The degree of elevation of bilirubin is much greater in small premature than in larger premature and full-term infants.

Since effects of Hykinone have been demonstrated in the glutathione system described above, this appears to be a likely mode of toxicity.[134] Glutathione levels are similar in premature and full-term infants.[152] The greater degrees of bilirubinemia noted in the smaller prematures may be related to a concomitant vitamin-E deficiency which allows greater damage to erythrocytes and/or to greater immaturity of the liver enzymes necessary for catabolism of bilirubin.[162]

It is of interest that toxic effects have not been noted in laboratory animals when the naturally occurring vitamin K_1 has been substituted; and also that vitamin K_1 has not produced an elevation of serum bilirubin in infants even when used in excessive amounts.[154, 155, 161] With respect to the water-soluble preparations, which include Synkayvite, menadione, and Hykinone as well as other preparations, it is unlikely that injections of 1 or 2 mg. will contribute significantly to the accumulation of bilirubin in infants with the "immaturity" defect in this system.[159, 161] However, even small amounts may cause a reaction in the "sensitive" infant with a genetically determined defect.

Anemia Related to Infection.—Both pallor and jaundice may represent clinical manifestations of sepsis in a young infant. Offending agents include the salivary gland virus, the parasite of toxoplasmosis, and the spirochete of syphilis as well as bacteria, especially of the coliform group.[29, 163-165]

Icterus was recently reported to have been present in 29 per cent of 87 patients under the age of 30 days in whom septicemia occurred.[163]

Kernicterus has occurred in a number of such infants.[29] It would seem that both accumulation of bilirubin in the serum and toxic changes in the blood-brain barrier and in cellular permeability permitting neural deposition of bilirubin might predispose to such a complication.

Since in the newborn period erythropoiesis is normally relatively inactive, it would follow that an unusually early onset of anemia must be due to the presence of some degree of increased destruction of erythrocytes.[24] Failure of the bone marrow to respond adequately to marked degrees of anemia does, of course, add to the severity and duration of the anemia. Infection may influence the rate of destruction of normal compatible donor cells as well as of the patient's own erythrocytes.[123] Whereas toxicity to the liver is a possible contributory factor in the accumulation of bilirubin, slight increases in hemolysis in the young infant with an immature liver are sufficient to explain the occurrence of jaundice.*

The most important aspect of this type of anemia lies in the recognition of the etiology and prompt treatment of the infection.

SUMMARY

A basic understanding of the consequences of hemolysis and of their intensification in infancy together with an awareness of the etiology and ultimate prognosis of each particular disorder is necessary to optimal management of infants in whom hemolytic disease occurs.

*The use of sulfonamides in the treatment of young infants in whom there is jaundice would seem contraindicated. Recent evidence suggests that these substances liberate bilirubin from protein-binding sites on serum proteins and may allow diffusion of bilirubin to other body compartments. Salicylates exert a similar effect and should therefore be considered hazardous in young infants with bilirubinemia.[167, 168]

REFERENCES

1. Brinkman, R., Wildshut, A., and Wittermans, A.: On the Occurrence of Two Kinds of Hemoglobin in Normal Human Blood, J. Physiol. **80:** 377, 1934.
2. Chernoff, Amoz I., and Singer, K.: Studies on Abnormal Hemoglobins. IV. Persistence of Fetal Hemoglobin in the Erythrocytes of Normal Children, Pediatrics **9:** 469, 1952.
3. Guest, G. M., and Brown, E. W.: Erythrocytes and Hemoglobin of the Blood in Infancy and Childhood. I. Size and Hemoglobin Content of the Erythrocytes in Nutritional Anemia, Am. J. Dis. Child. **52:** 616, 1936.
4. Whitby, L. E. H., and Britton, C. J. C.: Disorders of the Blood, ed. 6, Philadelphia, 1950, Blakiston, p. 120.
5. Pickles, M. M.: Hemolytic Disease of the Newborn, Oxford, 1949, Blackwell Scientific Publications.
6. Vaughan, V. C. III: Management of Hemolytic Disease of the Newborn, J. PEDIAT. **54:** 586, 1959.
7. Orlandini, O., Sass-Kortsak, A., and Ebbs, J. H.: Serum Gamma Globulin Levels in Normal Infants, Pediatrics **16:** 575, 1955.
8. Osborn, J. J., Dancis, J., and Julia, J. F.: Studies of Immunology of Newborn Infant, Pediatrics **9:** 736, 1952.
9. Janeway, C. A., and Gitlin, D.: The Gamma Globulins, Advances Pediat. **9:** 65, 1957.
10. Lemberg, R., and Legge, J. W.: Hematin Compounds and Bile Pigments, New York, 1949, Interscience Publishers.
11. Billing, B. H., and Lathe, G. H.: Excretion of Bilirubin as an Ester Glucuronide, Giving Direct van den Bergh Reaction, Biochem. J. **63:** 6P, 1956.
12. Schmid, R.: The Conjugation of Bilirubin With Glucuronic Acid, J. Lab. & Clin. Med. **48:** 940, 1956.
13. Levvy, C. A., and Storey, I. D. E.: Measurement of Glucuronide Synthesis by Tissue Preparations, Biochem. J. **44:** 295, 1949.
14. Karunairatnam, M. C., Kerr, L. M. H., and Levvy, C. A.: Glucuronide Synthesizing System in Mouse and Its Relationship to B-glucuronidase, Biochem. J. **45:** 496, 1949.
15. Grodsky, G. M., and Carbone, J. V.: Synthesis of Bilirubin Glucuronide by

Tissue Hemogenates, J. Biol. Chem. **226**: 449, 1957.
16. Schmid, R., Hammaker, L., and Axelrod, J.: Enzymatic Formation of Bilirubin Glucuronide, Arch. Biochem. **70**: 285, 1957.
17. Brown, A. K., and Zuelzer, W. W.: Studies on Neonatal Development of Glucuronide Conjugating System, J. Clin. Invest. **37**: 332, 1958.
18. Weech, A. A.: The Genesis of Physiologic Hyperbilirubinemia, Advances Pediat. **2**: 346, 1947.
19. Fashena, G. J.: Experimental Demonstration of Functional Hepatic Immaturity, Am. J. Dis. Child. **76**: 196, 1948.
20. Hsia, D. Y., Allen, F. H., Jr., Diamond, L. K., and Gellis, S. S.: Serum Bilirubin Levels in the Newborn Infant, J. PEDIAT. **42**: 277, 1953.
21. Zuelzer, W. W., and Kaplan, E.: ABO Heterospecific Pregnancy and Hemolytic Disease. III. Hematologic Findings and Erythrocyte Survival in Normal Infants, A. M. A. Am. J. Dis. Child. **88**: 307, 1954.
22. Obrinsky, W., Allen, E. L., and Anderson, E. E.: Physiologic Hyperbilirubinemia in Premature Infants, A. M. A. Am. J. Dis. Child. **87**, 305, 1954.
23. Brown, A. K., and Zuelzer, W. W.: Studies in Hyperbilirubinemia. I. Hyperbilirubinemia of the Newborn Unrelated to Isoimmunization, A. M. A. J. Dis. Child. **93**: 263, 1957.
24. Vaughan, V. C. III, Allen, F. H., Jr., and Diamond, L. K.: Erythroblastosis Fetalis. III. Prognosis in Relation to Clinical and Serologic Manifestations at Birth, Pediatrics **6**: 630, 1950.
25. Hsia, D., Allen, F. H., Jr., Gellis, S. S., and Diamond, L. K.: Erythroblastosis Fetalis. VIII. Studies of Serum Bilirubin in Relation to Kernicterus, New England J. Med. **247**: 668, 1952.
26. Claireaux, A. E., Cole, P. G., and Lathe, G. H.: Icterus of the Brain in the Newborn, Lancet **2**: 1226, 1953.
27. Waters, W. J., Richert, D., Rawson, H.: Bilirubin Encephalopathy, A. M. A. Am. J. Dis. Child. **86**: 483, 1953.
28. Meyers, T. C.: A Study of Serum Bilirubin Levels in Relation to Kernicterus and Prematurity, Arch. Dis. Childhood **31**: 75, 1956.
29. Zuelzer, W. W., and Mudgett, R. T.: Kernicterus: Etiologic Study Based on an Analysis of 55 Cases, Pediatrics **6**: 452, 1950.
30. Waters, W. J., and Britton, H. A.: Bilirubin Encephalopathy—Preliminary Studies Related to Production, Pediatrics **15**: 45, 1955.
31. Day, R.: Depression of Tissue Metabolism by Bilirubin, Hematin, and Mesobilirubin, A. M. A. Am. J. Dis. Child. **90**: 643, 1955.
32. Lathe, G. H.: Exchange Transfusion as a Means of Removing Bilirubin in Hemolytic Disease of the Newborn, Brit. M. J. **1**: 192, 1955.
33. Brown, A. K., Zuelzer, W. W., and Robinson, A. R.: Studies in Hyperbilirubinemia. II. Clearance of Bilirubin From Plasma and Extravascular Space in Newborn Infants During Exchange Transfusion, A. M. A. J. Dis. Child. **93**: 274, 1957.
34. Gairdner, D., Marks, J., and Roscoe, J. D.: Blood Formation in Infancy. I. Normal Bone Marrow, Arch. Dis. Childhood **27**: 128, 1952.
35. Gairdner, D., Marks, J., and Roscoe, J. D.: Blood Formation in Infancy. II. Normal Erythropoiesis, Arch. Dis. Childhood **27**: 214, 1952.
36. Robertson, S. E. J.: A Study of the Anemia Following Exchange Transfusion in Haemolytic Anemia of the Newborn, M. J. Australia **2**: 250, 1956.
37. Minkowski, O.: Ueber eine hereditäre, unter dem Bilde eines chronischen Ikterus mit Urobilinurie, Splenomegalie und Nierensiderosis verlaufende Affection, Verhandl. Deutsch. Kongr. f. inn. Med. **18**: 316, 1900.
38. Chauffard, A.: Pathogène de l'ictère congénital de l'adulte, Semaine méd. **27**: 25, 1907.
39. Meulengracht, E.: Ueber die Erblichkeitsverhältnisse beim chronischen hereditären hämolytischen Ikterus, Deutsches Arch. klin. Med. **136**: 33, 1921.
40. Campbell, J. M. H., and Warner, E. C.: Heredity in Acholuric Jaundice, Quart. J. Med. **19**: 333, 1926.
41. Debré, R., Lamy, M., Sée, G., and Schrameck, G.: Congenital and Familial Hemolytic Disease in Children, Am. J. Dis. Child. **56**: 1189, 1938.
42. Butterworth, C. E., Jr., Kracke, R. R., and Riser, W. H.: Hereditary Spherocytic Anemia in the Negro, Blood **5**: 793, 1950.
43. Shapiro, C. M., Josephson, A. M., Rozengvaig, S., and Kauffman, A.: Hereditary Spherocytosis in the Neonatal Period, J. PEDIAT. **50**: 308, 1957.
44. Haden, R. L.: The Mechanism of the Increased Fragility in Congenital Hemolytic Jaundice, Am. J. M. Sc. **188**: 441, 1934.
45. Selwyn, J. G., and Dacie, J. V.: Autohemolysis and Other Changes Resulting From the Incubation in Vitro of Red Cells From Patients With Congenital Hemolytic Anemia, Blood **9**: 414, 1954.
46. Motulsky, A. G., Giblett, E., Coleman, D., Gabrio, B., and Finch, C. A.: Life Span, Glucose Metabolism and Osmotic Fragility of Erythrocytes in Hereditary Spherocytosis, J. Clin. Invest. **34**: 911, 1955.

47. Prankerd, T. A. J., Altman, K. I., and Young, L. E.: Abnormalities of Carbohydrate Metabolism of Red Cells in Hereditary Spherocytosis, J. Clin. Invest. **34**: 1268, 1955.
48. Dacie, J. V., and Mollison, P. L.: Survival of Normal Erythrocytes After Transfusion to Patients With Familial Hemolytic Anemia, Lancet **1**: 550, 1943.
49. Dawson, L.: Hume Lectures on Hemolytic Icterus, Brit. M. J. **1**: 963, 1931.
50. Sharpe, J. C., Mc Laughlin, C. W., and Cunningham, R.: Hemolytic Jaundice: Immediate and Delayed Changes in the Blood After Splenectomy, Arch. Int. Med. **64**: 268, 1939.
51. Gänsslen, M., Zipperlen, E., and Schüz, E.: Die hämolytische konstitution, Deutsches Arch. klin. Med. **146**: 1, 1925.
52. Erlandson, M. E., Schulman, I., Smith, C. H.: Studies on Congenital Hemolytic Syndromes. II. Rates of Destruction and Production of Red Blood Cells in Hereditary Spherocytosis, Pediatrics (In Press).
53. Owren, P. A.: The Pathology of the "Hemolytic Crisis," Blood **3**: 231, 1948.
54. King, H., and Shumaker, H. B.: Splenic Studies. I. Susceptibility to Infection After Splenectomy Performed in Infancy, Ann. Surg. **136**: 239, 1952.
55. Gofstein, R., and Gellis, S. S.: Splenectomy in Infancy and Childhood: The Question of Overwhelming Infection Following Operation, A. M. A. J. Dis. Child. **91**: 566, 1956.
56. Smith, C. H., Erlandson, M. E., Schulman, I., and Stern, G.: Hazard of Severe Infections in Splenectomized Infants and Children, Am. J. Med. **22**: 390, 1957.
57. Robinson, G. C.: Hereditary Spherocytosis in Infancy, J. PEDIAT. **50**: 446, 1957.
58. Burman, D.: Congenital Spherocytosis in Infancy, Arch. Dis. Childhood **33**: 335, 1958.
59. Huntley, C. C.: Infection Following Splenectomy in Infants and Children, A. M. A. J. Dis. Child. **95**: 477, 1958.
60. Young, L. E., Izzo, M. J., and Platzer, R. F.: Hereditary Spherocytosis. I. Clinical Hematologic, and Genetic Features in 28 Cases With Particular Reference to Osmotic and Mechanical Fragility of Incubated Erythrocytes, Blood **6**: 1073, 1951.
61. Schiff, E.: Congenital Haemolytic Anemia With Normal Fragility of Red Blood Cells, Arch. Dis. Childhood **13**: 264, 1938.
62. Conrad, E., and Schmidt, R. E.: Congenital Hemolytic Anemia—A Case Requiring Early Splenectomy, Am. J. Dis. Child. **72**: 731, 1946.
63. Macauley, D.: Acholuric Jaundice in a Newborn Infant, Arch. Dis. Childhood **26**: 241, 1951.
64. Clinical Conference at St. Christopher's Hospital for Children, Philadelphia, Pa. Two Cases of Hereditary Spherocytosis Manifest in the Newborn Period, J. PEDIAT. **44**: 213, 1954.
65. Stenstrom, J. D., and Ford, H. S.: Hereditary Spherocytosis, Canad. M. A. J. **74**: 34, 1956.
66. Stamey, C. C., and Diamond, L. K.: Congenital Hemolytic Anemia in the Newborn: Relationship to Kernicterus, A. M. A. J. Dis. Child. **94**: 616, 1957.
67. Smith, I., and Franklin, A. W.: Meeting at Queen Charlotte's Maternity Hospital. Clinical Demonstration. IV. Congenital Familial Spherocytosis With Hyperbilirubinaemia Treated by Exchange Transfusion, Proc. Roy. Soc. Med. **50**: 707, 1957.
68. Hawksley, J. C.: Acholuric Family Jaundice in an Infant, Proc. Roy. Soc. Med. **27**: 942, 1934.
69. Diamond, L. K.: Congenital Hemolytic Anemia in Infancy and Childhood, M. Clin. North America **21**: 401, 1937.
70. Josephs, H. W.: Studies in Haemolytic Anaemia. I. Haemolysis Compensatory Regeneration and Erythroblastosis, Bull. Johns Hopkins Hosp. **62**: 25, 1938.
71. Abt, A. F.: Hemolytic Disease in Infants, Am. J. Dis. Child. **60**: 812, 1940.
72. Fallon, M.: Jaundice and Conditions Associated With this Phenomenon, J. PEDIAT. 23: 721, 1943.
73. David, J. K., and Minot, A. S.: Hemolytic Anemia in Infancy, Am. J. Dis. Child. **68**: 327, 1944.
74. Pinckney, C.: Familial Haemolytic Anemia, Proc. Roy Soc. Med. **39**: 696, 1946.
75. Newns, G.: Quoted by Burman.[58]
76. Evans, T. S., Waters, L. L., and Lowman, R. M.: Hypersplenism: Indications for Surgery, Connecticut M. J. **18**: 569, 1954.
77. Edwards, W. M., and Heaton, L. D.: Hypersplenism, Am. Pract. & Digest Treat. **6**: 387, 1955.
78. Field, C. E., and Tan, E. E.: Chronic Haemolytic Anaemia; A Report of Three Cases of Cooley's Anaemia and Five Cases of Acholuric Jaundice Seen in Malaya With a Discussion of Differential Diagnosis, M. J. Malaya **10**: 60, 1955.
79. Betke, K.: Icterus neonatorum gravis als Manifestation einer hereditären Spharocytose, Ztschr. Kinderh. **78**: 359, 1956.
80. Turman, C. M., Vaughan, V. C., and Shelly, R. M.: Incidence of Hemolytic Disease of the Newborn Due to A or B Incompatibility, Am. J. Obst. & Gynec. **71**: 885, 1956.
81. Crosby, W. H.: Hereditary Nonspherocytic Hemolytic Anemia, Blood **5**: 233, 1950.

82. Thompson, W. P.: Hemolytic Jaundice: Its Diagnosis, Behavior and Treatment, Bull. New York Acad. Med. **15:** 177, 1939.
83. Haden, R. L.: A New Type of Hereditary Hemolytic Jaundice Without Spherocytosis, Am. J. M. Sc. **214:** 255, 1947.
84. Kaplan, E., and Zuelzer, W. W.: Familial Nonspherocytic Hemolytic Anemia, Blood **5:** 811, 1950.
85. Feinberg, A. W., and Watson, J.: Nonspherocytic Hemolytic Anemia With Basophilic Stippling: Report of a Case in a Negro, Blood **6:** 357, 1951.
86. Motulsky, A. G., Crosby, W. H., and Rappaport, H.: Hereditary Nonspherocytic Hemolytic Disease: A Study of a Singular Familial Hemolytic Syndrome, Blood **9:** 749, 1954.
87. Bruton, O. C., Crosby, W. H., Motulsky, A. G.: Hereditary Nonspherocytic Hemolytic Anemia Presenting as Hemolytic Disease of the Newborn Infant, Pediatrics **13:** 41, 1954.
88. Dacie, J. V.: The Haemolytic Anaemia, New York, 1954, Grune and Stratton, p. 104.
89. Lipton, E. L.: Elliptocytosis With Hemolytic Anemia: The Effects of Splenectomy, Pediatrics **15:** 67, 1955.
90. Wyandt, H., Bancroft, P. M., and Winship, T. O.: Elliptic Erythocytes in Man, Arch. Int. Med. **68:** 1043, 1941.
91. Glenn, F., Cornell, G. N., Smith, C. H., and Schulman, I.: Splenectomy in Children With Idiopathic Thrombocytopenic Purpura, Hereditary Spherocytosis and Mediterranean Anemia, Surg. Gynec. & Obst. **99:** 689, 1954.
92. Wintrobe, M. M., Mathews, E., Pollack, R., and Dobyns, B. M.: A Familial Hemopoietic Disorder in Italian Adolescents and Adults, J. A. M. A. **114:** 1530, 1940.
93. Smith, C. H.: Detection of Mild Types of Mediterranean (Cooley's) Anemia, Am. J. Dis. Child. **75:**505, 1948.
94. Cooley, T. B., and Lee, P.: A Series of Cases of Splenomegaly in Children With Anemia and Peculiar Bone Changes, Tr. Am. Pediat. Soc. **37:** 29, 1925.
95. Smith, C. H., Sisson, T. R. C., Floyd, W. H., and Siegal, S.: Serum Iron and Iron-Binding Capacity in Children With Severe Mediterranean (Cooley's) Anemia, Pediatrics **5:** 799, 1950.
96. Liquori, A. M.: Presence of Foetal Haemoglobin in Cooley's Anemia, Nature, London **167:** 950, 1951.
97. Ellis, J. T., Schulman, I., and Smith, C. H.: Generalized Siderosis With Fibrosis of Liver and Pancreas in Cooley's (Mediterranean) Anemia, Am. J. Path. **30:** 287, 1954.
98. Whipple, G. H., and Bradford, W. L.: Racial or Familial Anemia of Children Associated With Fundamental Disturbances of Bone and Pigment Metabolism, Am. J. Dis. Child. **44:** 336, 1932.
99. Rich, A.: Studies on the Hemoglobin of Cooley's Anemia and Cooley's Trait, Proc. Nat. Acad. Sc. **38:** 187, 1952.
100. Chernoff, A. I.: Immunologic Studies of Hemoglobins. I. The Production of Anti-Hemoglobin Sera and Their Immunologic Characteristics, Blood **8:** 399, 1953.
101. Kaplan, E., and Zuelzer, W. W.: Erythrocyte Survival Studies in Childhood. I. Methods and General Observations, J. Lab. & Clin. Med. **36:** 511, 1950.
102. Sturgeon, P., and Finch, C. A.: Erythrokinetics in Cooley's Anemia, Blood **12:** 64, 1957.
103. Erlandson, M. E., Schulman, I., Stern, G., and Smith, C. H.: Studies of Congenital Hemolytic Syndromes. I. Rates of Destruction and Production of Red Blood Cells in Thalassemia, Pediatrics **22:** 910, 1958.
104. Schulman, I., Smith, C. H., and Stern, G.: Studies on the Anemia of Prematurity, A. M. A. Am. J. Dis. Child. **88:** 567, 1954.
105. Banton, A. H.: The Development of Mediterranean Anemia, Arch. Dis. Childhood **26:** 235, 1951.
106. Fawdry: Quoted by Banton.[105]
107. de Silva, C. C., and Weeratunge, C. E. S.: Cooley's Anaemia in Sinhalese Children, Arch. Dis. Childhood **26:** 224, 1951.
108. Baty, J. M., Blackfan, K. D., and Diamond, L. K.: Blood Studies in Infants and in Children. I. Erythroblastic Anemia; a Clinical and Pathological Study, Am. J. Dis. Child. **43:** 667, 1932.
109. Herrick, J. B.: Peculiar Elongated and Sickle-Shaped Red Corpuscles in a Case of Severe Anemia, Arch. Int. Med. **6:** 517, 1910.
110. Pauling, L., Itano, H. A., Singer, S. J., and Wells, I. C.: Sickle Cell Anemia, a Molecular Disease, Science **110:** 543, 1949.
111. Harris, J. W.: Studies on Destruction of Red Blood Cells. VIII. Molecular Orientation in Sickle Cell Hemoglobin Solutions, Proc. Soc. Exper. Biol. & Med. **75:** 197, 1950.
112. Singer, K., and Singer, L.: Studies on Abnormal Hemoglobins. VIII. The Gelling Phenomenon of Sickle Cell Hemoglobin: Its Biologic and Diagnostic Significance, Blood **8:** 1008, 1953.
113. Ingram, V. M.: Gene Mutations in Human Haemoglobin: The Chemical Difference Between Normal and Sickle Cell Haemoglobin, Nature **180:** 326, 1957.
114. Cooley, T. B., and Lee, P.: Sickle Cell Anemia in a Greek Family, Am. J. Dis. Child. **38:** 103, 1929.
115. Cooke, J. V., and Mack, J. K.: Sickle Cell Anemia in a White American Family, J. PEDIAT. **5:** 601, 1934.

116. Wells, I. C., and Itano, H. A.: Ratio of Sickle Cell Anemia Hemoglobin to Normal Hemoglobin in Sicklemics, J. Biol. Chem. **188:** 65, 1951.
117. Singer, K., Chernoff, A. I., and Singer, L.: Studies on Abnormal Hemoglobins. I. Their Identification in Sickle Cell Anemia and Other Hematologic Disorders by Means of Alkali Denaturation, Blood **6:** 413, 1951.
118. Singer, K., Motulsky, A. G., and Wile, S. A.: Aplastic Crisis in Sickle Cell Anemia. A Study of Its Mechanism and Its Relationship to Other Types of Hemolytic Crisis, J. Lab. & Clin. Med. **35:** 721, 1950.
119. Smith, C. H.: The Abnormal Hemoglobins: Clinical and Hematologic Aspects, J. PEDIAT. **50:** 91, 1957.
120. Watson, J.: The Significance of the Paucity of Sickle Cells in Newborn Negro Infants, Am. J. M. Sc. **215:** 419, 1948.
121. Schneider, R. G., and Haggard, M. E.: Sickling, a Quantitatively Delayed Genetic Character, Proc. Soc. Exper. Biol. & Med. **89:** 196, 1955.
122. Shields, G. S., Lichtman, H. C., Messite, J., and Watson, R. J.: Studies in Sickle Cell Disease. I. Quantitative Aspects of Sickling in the Newborn Period, Pediatrics **22:** 309, 1958.
123. Leiken, S. L., and McCoo, J. W., Jr.: Sickle-Cell Anemia in Infancy, A. M. A. J. Dis. Child. **96:** 51, 1958.
124. Frazier, C. A., and Rice, C. E.: Neonatal Sickle Cell Anemia, J. A. M. A. **143:** 1065, 1950.
125. Cohen, S. M., Bertram, W. M., and Orris, H. W.: Fatal Sickle Cell Anemia in a One-Month-Old Infant, J. PEDIAT. **30:** 468, 1947.
126. Trowell, H. C., Raper, A. B., and Welbourn, H. F.: The Natural History of Homozygous Sickle-Cell Anaemia in Central Africa, Quart. J. Med. **26:** 401, 1957.
127. MacIver, J. E., Went, L. N., and Cruickshank, E. K.: Sickle Cell-Thalassemia Disease in Jamaica, Blood **13:** 359, 1958.
128. Wollstein, M., and Kreidel, K. V.: Sickle Cell Anemia, Am. J. Dis. Child. **36:** 998, 1928.
129. McGavack, T. H., and Nussbaum, C. C.: Skin Manifestations of Sickle Cell Anemia, Urol. & Cutan. Rev. **46:** 194, 1942.
130. Gasser, C.: Aplasia of Erythropoiesis, Pediat. Clin. North America, May, 1957, p. 445.
131. Zuelzer, W. W., and Apt, L.: Acute Hemolytic Anemia Due to Naphthalene Poisoning, J. A. M. A. **141:** 185, 1949.
132. Schafer, W. B.: Acute Hemolytic Anemia Related to Naphthalene. Report of a Case in a Newborn Infant, Pediatrics **7:** 172, 1951.
133. Cock, T. C.: Acute Hemolytic Anemia in the Neonatal Period, A. M. A. J. Dis. Child. **94:** 77, 1957.
134. Zinkham, W. H., and Childs, B.: Effect of Vitamin K and Naphthalene Metabolites on Glutathione Metabolism of Erythrocytes From Normal Newborns and Patients With Naphthalene Hemolytic Anemia (abstr.), A. M. A. J. Dis. Child. **94:** 420, 1957.
135. Zinkham, W. H., and Childs, B.: A Defect Glutathione Metabolism in Erythrocytes From Patients With a Naphthalene-Induced Hemolytic Anemia, Pediatrics **22:** 461, 1958.
136. Mackell, J. V., Reiders, F., Brieger, H., and Breuer, E. L.: Acute Hemolytic Anemia Due to Ingestion of Moth Balls. I. Clinical Aspects. II. Hemolytic Action of Naphthalene and Its Oxidation Products, Pediatrics **7:** 722, 1951.
137. Chusid, E., and Fried, C. T.: Acute Hemolytic Anemia Due to Naphthalene Ingestion, A. M. A. Am. J. Dis. Child. **89:** 612, 1955.
138. McGovern, J. J., Isselbacher, K., Rose, P. J., and Grossman, M. S.: Observations on the Glutathione (GSH) Stability of Red Blood Cells, A. M. A. J. Dis. Child. **96:** 502, 1958.
139. Smillie, W. G.: B-Naphthol Poisoning in the Treatment of Hookworm, J. A. M. A. **74:** 1503, 1920.
140. Hockwald, R. S., Arnold, J., Clayman, C. B., and Alving, A. S.: Status of Primaquine. IV. Toxicity of Primaquine in Negroes, J. A. M. A. **149:** 1568, 1952.
141. Derm, R. J., Beutler, E., and Alving, A. S.: The Hemolytic Effect of Primaquine. V. Primaquine as a Manifestation of a Multiple Drug Sensitivity, J. Lab. & Clin. Med. **45:** 30, 1955.
142. Dern, R. J., Weinstein, I. M., Le Roy, G. V., Talmadge, D. W., and Alving, A. S.: The Hemolytic Effect of Primaquine. I. The Localization of the Drug-Induced Hemolytic Defect in Primaquine-Sensitive Individuals, J. Lab. & Clin. Med. **43:** 303, 1954.
143. Beutler, E., Dern, R. J., and Alving, A. S.: The Hemolytic Effect of Primaquine. III. A Study of Primaquine-Sensitive Erythrocytes, J. Lab. & Clin. Med. **44:** 177, 1954.
144. Dern, R. J., Beutler, E., and Alving, A. S.: The Hemolytic Effect of Primaquine. II. The Natural Course of the Hemolytic Anemia and the Mechanism of Its Self-Limited Character, J. Lab. & Clin. Med. **44:** 171, 1954.
145. Beutler, E., Dern, R. J., and Alving, A. S.: The Hemolytic Effect of Primaquine. IV. The Relationship of Cell Age to Hemolysis, J. Lab. & Clin. Med. **44:** 439, 1954.
146. Beutler, E., Dern, R. J., Flanagan, C. L., and Alving, A. S.: The Hemolytic Effect of Primaquine. VII. Biochemical

Studies of Drug-Sensitive Erythrocytes, J. Lab. & Clin. Med. **45**: 286, 1955.

147. Beutler, E.: The Glutathione Instability of Drug-Sensitive Red Cells, J. Lab. & Clin. Med. **49**: 84, 1957.
148. Carson, P. E., Flanagan, C. L., Ickes, C. E., Alving, A. S.: Enzymatic Deficiency in Primaquine-Sensitive Erythrocytes, Science **124**: 484, 1956.
149. Childs, B., Zinkham, W. H., Browne, E. A., Kimbro, E. L., and Torbert, J. V.: A Genetic Study of a Defect in Glutathione Metabolism of the Erythrocyte, Bull. Johns Hopkins Hosp. **102**: 21, 1958.
150. Gross, R. T., Marks, P. A., Hurwitz, R. E., Beasley, T., and Sui, M.: Studies on the Hereditary Enzymatic Defect in Certain Drug-Induced Hemolytic Anemias, A. M. A. J. Dis. Child. **96**: 436, 1958.
151. Newton, W. A., Jr., and Bass, J. C.: Glutathione-Sensitive Chronic Nonspherocytic Hemolytic Anemia, A. M. A. J. Dis. Child. **96**: 501, 1958.
152. Scheinberg, A., Ramot, B., Adam, A., and Sheba, Ch.: Glutathione Metabolism in Cord Blood and the Newborn Infant, A. M. A. J. Dis. Child. **96**: 542, 1958.
153. Gross, R. T., and Hurwitz, R. E.: The Pentose Phosphate Pathway in Human Erythrocytes: Relationship Between Age of the Subject and Enzyme Activity, Pediatrics **22**: 453, 1958.
154. Molitor, H., and Robinson, H. J.: Oral and Parenteral Toxicity of Vitamin K_1, Phthiocol and 2 Methyl 1,4, Naphthoquinone, Proc. Soc. Exper. Biol. & Med. **43**: 125, 1940.
155. Ansbacher, S., Corwin, W. C., and Thomas, B. G. H.: Toxicity of Menadione, Menadiol, and Esters, J. Pharmacol. **75**: 111, 1942.
156. Gasser, C.: Die hämolytische frühgeburtenanämie mit spontaner Innenkörperbildung, Helvet. paedit. acta **8**: 491, 1953.
157. Allison, A. C.: Danger of Vitamin K to Newborn, Lancet **1**: 669, 1955.
158. Laurence, B.: Danger of Vitamin-K Analogues to Newborn, Lancet **1**: 819, 1955.
159. Bound, J. P., and Telfer, T. P.: Effect of Vitamin-K Dosage on Plasma-bilirubin Levels in Premature Infants, Lancet **1**: 720, 1956.
160. Meyer, T. C., and Angus, J.: The Effect of Large Doses of "Synkavit" in the Newborn, Arch. Dis. Childhood **31**: 212, 1956.
161. Asteriadon-Samartzis, E., and Leiken, S.: The Relation of Vitamin K to Hyperbilirubinemia, Pediatrics **21**: 397, 1958.
162. Allison, A. C., Moore, T., and Sharman, I. M.: Haemolysis and Haemoglobinuria in Vitamin-E Deficient Rats After Injections of Vitamin-K Substitutes, Brit. J. Haemat. **2**: 197, 1956.
163. Smith, R. T., Platou, E. S., and Good, R. A.: Septicemia of the Newborn: Current Status of the Problem, Pediatrics **17**: 549, 1956.
164. Margileth, A. M.: The Diagnosis and Treatment of Generalized Cytomegalic Inclusion Disease of the Newborn, Pediatrics **15**: 270, 1955.
165. Eichenwald, H.: Congenital Toxoplasmosis, Copenhagen, 1958, Special Publication of International Pediatric Congress (In Press).
166. Guest, G. M., and Browne, E. W.: Erythrocytes and Hemoglobin of the Blood in Infancy and Childhood. III. Factors in Variability Statistical Studies, A. M. A. J. Dis. Child. **93**: 486, 1957.
167. Odell, G. B.: In Vitro Studies of the Effect of Sulfonamides on Bilirubin, A. M. A. J. Dis. Child. **96**: 535, 1958.
168. Ibid.: Comment; Transactions of Society for Pediatric Research, A. M. A. J. Dis. Child. **96**: 441, 1958.

MANAGEMENT OF HEMOLYTIC DISEASE OF THE NEWBORN INFANT

VICTOR C. VAUGHAN III, M.D.
AUGUSTA, GA.

THE management of hemolytic disease of the newborn infant requires familiarity with, and consideration of, three major problems which the affected infant may present. The first is the *threat of stillbirth;* the second is the immediate threat to adequate resuscitation or long life presented by *severe illness at the moment of birth;* and the third is the *prevention of kernicterus.* While these three problems are undeniably related to each other as manifestations of severe hemolytic disease, each demands quite different orientation to the immediate needs of the infant and each deserves to be handled separately as a special problem of management of infants with hemolytic disease.

CASE FINDING

Whatever may be the nature of the immediate threat to the survival of the infant with hemolytic disease, management begins with *case finding,* the nature of which is generally familiar. The antenatal determination of the Rh type of the pregnant woman is essential and should be carried out in every woman in every pregnancy. Clerical and technical errors are too common to permit last year's report to stand for all time. Whenever a sample of blood submitted ante partum proves to be that of an Rh-negative (D-negative) woman, it should be examined for anti-Rh (anti-D) antibody. If no antibody is found in a specimen submitted early in pregnancy, the outlook is generally favorable, but a second specimen should be tested at about 35 to 36 weeks' gestation. Absence of antibody at that time renders the possibility of significant degree of hemolytic disease very remote.

These two tests for antibody in the serum of the Rh-negative mother, one early and one late in pregnancy, constitute *minimal* adequate care. While it is fashionable in many clinics to examine maternal serum for antibody at more or less regular or increasingly frequent intervals during pregnancy, even when an early test shows no sensitization, this procedure may be as often productive of unnecessary anxiety as of any real reassurance in the 19 out of 20 Rh-negative women who will never have an infant with hemolytic disease. Moreover, any possible value inherent in serial determinations of titer in *sensitized* women is likely to be realized only when the practice in the laboratory is to save frozen samples of serum from all previous specimens for re-examination with each new specimen, using the same test cell, incubation, and the like. Changes in titer which are not so accurately controlled are of very doubtful help in the management of pregnancy complicated by isoimmunization.

From the Department of Pediatrics, Medical College of Georgia.

If antibody is found in the Rh-negative mother early in pregnancy, the possibility of hemolytic disease in the infant becomes large, presuming the father of the infant to be Rh positive, and the risk of intrauterine death considerable. If antibody is found to appear during pregnancy between the earlier and the later tests, the likelihood of stillbirth or of severe disease is not quite so great, but plans must be made to safeguard the remainder of the pregnancy and the neonatal period. It seems unnecessary to determine the father's Rh type except when antibody is found in maternal serum, at which time his probable genotype should be established.

Certain other mothers will need the same attention as the Rh-negative mother whose case-finding instrument in hemolytic disease was antenatal testing. These will include all women who have ever in the past had a blood transfusion or an injection of intramuscular blood and those women who have had stillbirths which were not explained or infants displaying conspicuous icterus or anemia in the neonatal period. For these mothers, as for Rh-negative mothers, adequate serologic study demands that their serums be tested against all antigens within the Rh system and against those other blood antigens, such as Kell, Duffy, S, s, Kidd, and the like, which may have been stimulated by transfusions received or may have been responsible for observed difficulties. The red blood cells of the husband should also be used as test cell whenever possible.

In the cases of the Rh-negative mother who has no antibody and of Rh-positive mothers who are not tested because they have not received any blood transfusion, give no history of suspicious illness in a previous child, and have had uneventful pregnancies and deliveries, case finding in respect to hemolytic disease consists of close observation of the newborn infant at the time of birth for signs of illness and continued observation during the first few days of life for the *early onset of icterus* or for its conspicuous *intensity*. Any newborn infant seen to become icteric within the first 24 to 36 hours of life or who has more than mild icterus at any time thereafter deserves *immediate* careful study for the possibility that hemolytic disease may be present.

THE THREAT OF STILLBIRTH

The threat of stillbirth most commonly exists in Rh incompatibility. For practical purposes, stillbirth does not occur in A or B incompatibility, and those sensitizations such as in the Kell system or S system which might give rise to stillbirth owing to their sometimes greater severity are quite rare.

The likelihood of stillbirth as a result of Rh incompatibility is measurably related statistically to the level of maternal antibody, with high levels carrying increased risk. Unfortunately, while the statistical relationship cannot be denied, often individual variations in titer are of such nature as to render the serial determination of antibody titer an unsatisfactory instrument by which to measure the immediate threat of stillbirth in any given instance. Of more significance than consideration of titer is the observation that, irrespective of titer, the probability of stillbirth becomes very high in instances where a

mother has had a previously stillborn infant as a result of hemolytic disease.

No method is currently known whereby hemolytic disease can be ameliorated so long as the infant remains in utero. A variety of nutritional and endocrine products have been given to sensitized mothers without convincing evidence of benefit. No Rh hapten has confidently been shown to exist. Under these circumstances, the possibility of preventing a stillbirth which must otherwise occur before the spontaneous delivery of the infant resides in the early delivery of the infant, not only before death occurs but before the baby becomes so ill that he cannot survive though liveborn.

Between 1948 and 1952, it was shown with confidence[1, 2] that the early delivery of the infant with hemolytic disease carried a substantially greater risk of his death or injury from kernicterus. For this reason, early delivery of the potentially affected infant was generally discouraged. Within the past three to four years, however, it has become apparent that *multiple* exchange transfusions have considerable promise in the prevention of kernicterus even in premature infants. The possibility is therefore being re-explored that early delivery of the threatened infant may in some measure answer the problem of stillbirth in hemolytic disease.

The data of Allen,[3] which clearly depict the natural history of stillbirth in hemolytic disease, offer essential background against which to view the difficulties of a program of prevention of stillbirth which has the highest probability of being helpful in any significant degree. Allen observed that in 174 sensitized Rh-negative mothers with Rh-positive fetuses in whom titers of anti-Rh antibody remained less than 1:64 during pregnancy and who had no history of previous stillbirth, the cumulative rate of stillbirth was 2 per cent at 33 weeks' gestation, 4 per cent at 35 weeks, and 8 per cent at 40 weeks; 4 per cent of those infants alive at 37 weeks' gestation were eventually stillborn. In 186 sensitized mothers in whom anti-Rh titers reached 1:64 or higher, without a past history of stillbirth, the cumulative rate of stillbirth was 10 per cent by 33 weeks, 16 per cent by 35 weeks, 17 per cent by 37 weeks, and 29 per cent by 40 weeks; 14 per cent of infants alive at 37 weeks' gestation were subsequently stillborn. In 25 women in whom a titer below 1:64 was recorded but who gave a history of stillbirth, 25 per cent of fetuses were lost by 33 weeks, 32 per cent by 35 weeks, 40 per cent by 37 weeks; 1 of 15 infants alive at 37 weeks' gestation was later lost. By contrast, in 59 women who had both anti-Rh titers of 1:64 or higher and a previous stillbirth, 34 per cent of infants were stillborn by 33 weeks' gestation, 46 per cent by 35 weeks, 68 per cent by 37 weeks, and 78 per cent by 40 weeks; 65 per cent of infants alive at 33 weeks were subsequently stillborn; 59 per cent of those alive at 35 weeks and 32 per cent of those still alive at 37 weeks were stillborn.

These data show the unmistakable relationship of stillbirth to maternal reproductive history and to titer. They make it certain, moreover, that if a substantial number of infants expected to be stillborn are to be saved through early delivery of mothers with high titers and who have had previously stillborn infants, then delivery must be planned for considerably earlier

than the thirty-seventh week of pregnancy. In view of the still experimental and exploratory nature of this current re-examination of the potentialities of early delivery in management of hemolytic disease, it is impossible to give firm criteria by which patients who may profit from this form of management may be chosen or how the optimal time and method of delivery may be selected. Chown and Bowman[4] have selected 33 to 34 weeks as the time when such mothers may with some measure of success begin to be delivered. They have suggested that medical induction of labor should be undertaken wherever possible, though delivery by cesarean section as early as 32 to 33 weeks has, exceptionally, lead to the survival of infants who might otherwise have suffered intrauterine death. Allen has had less satisfactory results in a small number of patients delivered at 35 weeks by cesarean section than Chown has had with medical induction of labor and pelvic delivery.

The mother at highest risk is quite clearly the mother who has already demonstrated in an earlier pregnancy her capacity for producing a stillborn infant. If she has already given birth to two stillborn infants, her likelihood of having a satisfactory result in a succeeding pregnancy without medical intervention probably does not exceed one chance in ten. When such a mother can be shown to have a homozygous Rh-positive husband, she would appear to be a good candidate for early delivery in spite of the increased hazard to the infant from prematurity and from kernicterus.

In instances where the sensitized mother has a heterozygous husband or where she gives no history of a previously affected infant and her titer remains less than 1:64, there would seem to be little reason for interruption of pregnancy. On the other hand, Allen has suggested that in mothers with rises of antibody titer to high levels in the pregnancies which are the first known to be complicated by sensitization, there may be some virtue in more or less routine delivery at about 37 weeks if *medical* induction of labor is possible. He does not feel that these circumstances warrant delivery by cesarean section.

Careful attention to the subjective impressions of the mother who is at high risk may help in the selection of optimal time for her early delivery. Many such mothers have a heightened sense of discomfort which appears closely related in time to the rapidly developing serious illness of the fetus or impending intrauterine death. The symptoms are a feeling of unduly rapid enlargement, increased pressure, and diminished fetal movement; often there are objective findings of polyhydramnios, but occasionally objective obstetric correlates of these maternal complaints are not found, even when intrauterine death is imminent. In the mother who has previous experience of these sensations, her own impression of the earliest such sign of fetal distress may point to the appropriate time for immediate delivery.

The warning that early delivery of the sensitized Rh-negative mother should be contemplated only where the risk of stillbirth is great cannot be made too strong, inasmuch as this uncertain solution to the problem of stillbirth leads directly to and intensifies the problem of prevention of kernicterus. Under no circumstances

should early delivery be undertaken unless it is certain that an experienced team is on hand at the time of delivery who can adequately handle the problems of the severely ill infant and who will be able to carry out multiple exchange transfusions if these should be necessary for the prevention of kernicterus.

It is among the sensitized mothers of severely ill and stillborn infants that the rare complication of hypofibrinogenemia may be encountered. Serial determinations of the fibrinogen content of plasma may be indicated; occasionally the threat of developing afibrinogenemia may call for early delivery of such a mother.

THE DELIVERY OF THE INFANT OF THE SENSITIZED MOTHER

At whatever time in pregnancy or by whatever method the delivery of the sensitized mother is planned, the circumstances surrounding birth of the infant ought to include a state of preparedness for urgently needed treatment of the infant. The delivery should be managed with as little analgesia or anesthesia as possible, since infants with anemia due to hemolytic disease may tolerate analgesic or anesthetic agents poorly. Delivery without drugs is ideal, where this is possible; the preferred anesthetic agents will be local rather than general.

Allen has summarized the argument for *planned* delivery of the sensitized mother: It can be carried out at a time when the necessary personnel and equipment can be made most conveniently and effectively available for the treatment of the potentially ill infant. In general, however, it will likely be found best in most situations to permit pregnancy to terminate spontaneously.

It will be wise, in general, to clamp the umbilical cord as soon as the delivery of the infant permits this, since severely ill infants may have an increase in blood volume and in venous pressure. The cause of these findings is uncertain, but it is probably a compound of anemia, hypoproteinemia, anoxia, and cardiac failure. If the umbilical flow is not promptly interrupted, the infant with advanced hemolytic disease may be made more dangerously ill, owing to that increase in blood volume which will result from transfusion of blood from the placenta. Infants who are *not* severely ill will generally not suffer important adverse consequences of early interruption of the cord. An exceptional situation would occasionally exist where evidence was at hand that the infant might have lost blood by retroplacental or other hemorrhage prior to delivery, such as has been described by Chown.[5]

Careful examination of the infant of the sensitized mother at the time of birth is of utmost importance. It is sometimes easy to see in the pale, limp, edematous, or bloated infant the urgent need for immediate therapy; on the other hand, other infants whose need for treatment is scarcely less immediate may present no outstanding sign of illness at the moment of birth. It has been repeatedly emphasized that jaundice is almost never present at the moment of birth. On the other hand, in those infants in whom the hemolytic process has been particularly active, there is likely to be enlargement of both liver and spleen, sometimes accompanied by mild edema of the extremities, and pallor which may be obscured by blood, mucus, vernix, or mild cyanosis. Chown has emphasized that the umbilical cord of the infant

with severe hemolytic disease often displays mild yellowish discoloration and is likely to show venous congestion with vigorous pulsations. The enlarged liver and spleen may be congested and friable; rupture of these organs with fatal hemorrhage may be the result of improper or ungentle handling of the infant. The possibility that such may have occurred should always be entertained in the infant suspected of hemolytic disease who appears to be in shock.

Laboratory studies for the purpose of confirmation of the diagnosis of hemolytic disease are scarcely necessary in the infant born to the sensitized mother who presents such signs of severe illness as those enumerated. Occasionally the infant's needs for therapy will be so urgent that such tests should be put off until the immediate condition of the infant can be improved. The majority of infants, however, who are liveborn to sensitized mothers will present considerably milder or no clinical signs of illness. In these infants an initial examination will establish what findings, if any, exist to suggest hemolytic disease, and decisions as to the infant's need for therapy will generally rest upon the results of studies performed on cord blood or of serial studies of the infant's blood during the next few hours.

In the infant born to the sensitized mother, the variety of studies carried out on cord blood for confirmation of illness or estimation of its severity may be large or small, but they *must* include determination of the *infant's Rh type, hemoglobin level,* and level of *serum bilirubin.* Demonstration that the infant of the mother who has anti-Rh antibodies in her serum is Rh positive establishes the diagnosis of hemolytic disease of the newborn infant. Other clinical or laboratory evidence of disease may not be present, but the direct antiglobulin test (Coombs test) is so positive in such infants (and often strongly so) that a negative test suggests a laboratory error with far more likelihood than it suggests any such possibilities as that the infant is truly negative or has mild disease or does not have a potentially serious disease. We have observed kernicterus in infants in whom it occurred only because an optimistic view was taken of a negative Coombs test. This negative test has most often proved to be a laboratory error but in two instances appeared consistently negative with repeated examination. A positive test, then, merely confirms the diagnosis of hemolytic disease while a negative test may not exclude it. There is one circumstance, on the other hand, where the direct antiglobulin test is of great value and absolutely essential; that is, when the offspring of a sensitized Rh-negative mother appears to be Rh negative to direct test with anti-Rh typing serum. If such an infant proves to have a positive Coombs test, he should be regarded as Rh positive until some other explanation for this positive reaction can be found. Usually the meaning of this reaction will be that the infant's cells have been coated by anti-Rh antibodies which have blocking activity. Such infants will generally need active therapy.

The determination of hemoglobin level in cord blood gives a very helpful estimate of the severity of the hemolytic process. Other determinations such as the red blood cell count, the hematocrit, reticulocyte count, estimation of normoblastemia, and the like may add

completeness or elegance to the study of the infant but are likely to be less easily and less accurately measured than the hemoglobin level and add little to the basis on which estimation must be made of the need of the infant for treatment. It still appears not to be widely enough appreciated that after the opportunity to examine the hemoglobin level in cord blood is past, *the hemoglobin level offers NO help in the estimation of the need of the infant for exchange tranfusion for the prevention of kernicterus.*

The level of serum bilirubin in cord blood, like the hemoglobin, is related to the severity of the intrauterine hemolytic process but measures this severity less directly than does the hemoglobin level. Following delivery, there is for most practical purposes a dissociation of the hemoglobin level from the level of serum bilirubin, such that the hemoglobin level indicates only the need for such transfusion as will correct anemia, while the bilirubin level becomes the best available indication of the need for treatment to prevent kernicterus. This distinction is of the utmost importance, since treatment for anemia is adequately accomplished by small transfusions of packed cells whereas the prevention of kernicterus depends upon systematic employment of exchange transfusion.

IMMEDIATE TREATMENT OF HEMOLYTIC DISEASE PRESENTING SEVERE MANIFESTATIONS AT BIRTH

Some of the manifestations of hemolytic disease in infants severely affected at the moment of birth have been presented above. To these, we may add that petechial hemorrhages are not uncommon; in the majority of instances they apppear to represent anoxia, but in exceptional cases they may reflect a thrombocytopenia of isoimmunologic origin. The resuscitation of the infant severely ill at the moment of birth may present considerable difficulty, owing to the embarrassment of respiration by edema, ascites, or other factors associated with heart failure, increased blood volume, and elevation of venous pressure. In rare instances ascites may dominate the clinical picture and be appropriately handled by immediate paracentesis. More often, the severely ill infant will need nothing so urgently as to begin to have immediate relief of his excess of blood volume and restoration of the oxygen-carrying capacity of his blood. Wheeler and Ambuel[6] have clearly pointed out the goals which ought to be set in the treatment of such infants: There is no urgent need for immediate complete replacement of the infant's blood by means of exchange transfusion which may be unduly prolonged and poorly tolerated, but the immediate goal is to stabilize the infant's circulatory and respiratory status. Mollison and Cutbush[7] were the first to show that the venous pressure in the normal newborn infants rarely exceeds 8 cm. but that in these severely ill infants with hemolytic disease it may exceed 15 cm. or more. This pressure is most easily measured from the level of the abdominal skin at the umbilical region in a column of blood held within the tube through which an exchange transfusion is to be carried out. Wheeler and Ambuel point out that in the treatment of infants with elevated venous pressure it may make good sense to use a concentrate of sedimented cells in order to achieve the most satisfactory restoration of hemoglobin level with

the least prolongation of the transfusion procedure. It will often be found that whole blood will do as well, judiciously used.

The technique of exchange transfusion has been adequately described elsewhere[8] and will be presumed familiar. It will scarcely ever be necessary to use other than the umbilical route. By this route, in severely ill babies, 20 ml. of blood may be withdrawn and replaced by 10 to 15 ml. of donor blood, serially, until the venous pressure has been reduced from its elevated level to the neighborhood of 7 to 8 cm. This will occasionally require the withdrawal of as much as 50 to 70 ml. more blood than is introduced and may on rare occasions require an even more considerable exsanguination of the infant (up to 100 ml.). This procedure may take courage but, as Wheeler and Ambuel have stated, few more rewarding therapeutic results will be enjoyed by the physician than to see the gasping, pale, edematous infant with weak movements and a plaintive grunting cry restored so quickly to normal color and activity.

The results of exchange transfusion with diminution of the blood volume of the severely affected infant are not always dramatic, nor does every infant with severe anemia need such attention to his blood volume. Some infants remain in a precarious state following this procedure, even when they appear to have been greatly helped, and will warrant careful watching for the next few hours. In some it may be wise to reinsert the umbilical cannula for new measurements of the venous pressure at hourly or two-hourly intervals to make sure that a secondary rise in venous pressure has not accompanied the redistribution of body fluids. If the infant with severe disease is to survive, he will rarely, with adequate treatment, remain for more than 12 hours in the precarious state in which he was found at birth.

At whatever point the infant born with severe hemolytic disease seems well stabilized so far as respiratory and cardiac function is concerned, he enters a new phase of disease, wherein the threat is not early death from anemia and heart failure but death or disability associated with kernicterus. In this respect, following his early treatment, the infant with severe disease presents the same problem as the infant with a less severe hemolytic process who presented no urgent need for therapy at the moment of birth. In particular, further need for adjustment of blood volume usually ceases to exist.

PREVENTION OF KERNICTERUS

There is abundant evidence that kernicterus is the only serious threat to the survival of infants with hemolytic disease who are born in a satisfactory condition or who are brought into a satisfactory clinical state through the use of such early treatment as that described above. The evidence is good also that properly employed exchange transfusion will prevent kernicterus in most infants with hyperbilirubinemia if the bilirubin level can be maintained below 20 mg. per 100 ml. No other secure prophylactic measure is at hand. Such other possible aids to the prevention of kernicterus as ACTH, adrenal steroids, or glucuronic acid are at present of no proved value, deserving as they may be of further study. Such measures will be dangerous if they divert attention from the fact that kernicterus can be prevented

with exchange tranfusion, which we must regard as the *conservative* form of treatment of hemolytic disease or of hyperbilirubinemia.

Wheeler and Ambuel have very clearly pointed out the value and dangers of exchange transfusion, along with what it can and cannot be expected to accomplish in hemolytic disease. Moreover, they have pointed out some of the difficulties which attend the decision as to whether an exchange transfusion is or is not to be done in any given infant. Generally speaking, the following considerations will govern the decision as to whether treatment by exchange transfusion is carried out: (1) Infants with levels of serum bilirubin above 20 mg. per 100 ml. begin to be at high risk of kernicterus. (2) In infants with hemolytic disease, a rapidly rising level of serum bilirubin sustained over a period of several hours may, according to the observed rate, predict a level within a day or two which will exceed 20 mg. per 100 ml. (3) The bilirubin level in cord serum is a reflection of the intensity of the hemolytic process, of the rapidity with which the infant is forming bilirubin, and of the effectiveness with which the placenta is carrying on a continous process of removal of bilirubin from the baby's body fluids into the maternal circulation. Wheeler and Ambuel have stated that where the level of bilirubin in cord blood exceeds 4 mg. per 100 ml., future need for exchange transfusion to control an established or threatened level of serum bilirubin above 20 mg. per 100 ml. will be found in 80 per cent of infants; by contrast, where the level of serum bilirubin in cord blood is less than 4 mg. per 100 ml., such need for exchange transfusion will occur in 20 per cent of infants with hemolytic disease.

It is unlikely that any two observers with wide experience of hemolytic disease have the same criteria for election of exchange transfusion for the prophylaxis of kernicterus. We have accepted the following criteria: (1) The presence of clinical findings of hemolytic disease at birth, in the form of hepatosplenomegaly of conspicuous degree, pallor, or edema. Such infants are unlikely to handle hemolytic disease without need, sooner or later, for transfusion. They have an active hemolytic process, and exchange transfusion is regarded as valuable prophylaxis under these circumstances even when the threat of kernicterus seems for the moment remote. (2) A level of hemoglobin in cord blood below 14 Gm. per cent. Anemia, like clinical evidence of active disease, indicates an active hemolytic process and predicts a need for transfusion sooner or later, which may well be given now with the added positive value of prophylaxis against hyperbilirubinemia. (3) A level of bilirubin in cord serum which exceeds 3.5 mg. per 100 ml. The conspicuous elevation of serum bilirubin above the normal average level of 1.8 mg. per 100 ml. is taken to be predictive of ultimate rises in excess of 20 mg. per 100 ml.

In the infant with otherwise equivocal indications for exchange transfusion, *prematurity* by two weeks or more or history of severe disease in a previous child will be strong arguments for the prophylactic employment of exchange transfusion and may sometimes be felt to be compelling indications.

If none of the above indications exists for exchange transfusion at the

moment of birth, the infant may be followed carefully with serial bilirubins to determine whether a level of 20 mg. per 100 ml. has been exceeded or is likely to be exceeded within the near future. Since the level of serum bilirubin generally rises steadily in normal newborn infants until the third or fourth day of life, it seems appropriate to regard a rate of rise during the first day in excess of from 0.25 to 0.33 mg. per 100 ml. per hour as indicating the ultimate need for exchange transfusion to control a level which will exceed 20 mg. per 100 ml. Zuelzer and Cohen[9] have suggested that exchange transfusion should be done if the level of serum bilirubin exceeds 10 mg. per 100 ml. in the first 24 hours, 14 mg. per 100 ml. in the second 24 hours, or 17 mg. per 100 ml. at any time thereafter within the first four days of life. We have accepted a commitment to keep the level of serum bilirubin below 20 mg. per 100 ml. during the first five days of life in full-term infants and for a longer period in premature infants, since the peak levels of bilirubin in the premature infant occur somewhat later than in the full-term infant.

Certain practical points regarding the dynamics of the removal of bilirubin through exchange transfusion deserve to be very clearly understood if this procedure is to be used most effectively and appropriately for the control of hyperbilirubinemia. First, in an infant of average size (3 kilograms), an exchange transfusion which employs a normal amount of blood (500 ml.) can be expected to remove about 85 per cent of the infant's original red blood cell mass. However, it will lower the level of serum bilirubin to only approximately ⅓ to ¼ of the original value and will remove only about ¼ of the total amount of bilirubin which is contained in body fluids. Following the exchange transfusion, as Brown, Zuelzer, and Robinson[10] have shown, there is a very prompt restoration of bilirubin toward the original value, and this restoration generally attains about 75 per cent of the original value. This phenomenon is clearly the result of redistribution of bilirubin in body fluids, and this takes place surprisingly rapidly. The redistribution in a small number of infants studied by us[11] appears to be virtually complete at the end of one hour or a little more.

There are two important consequences of the fact that only a small proportion of the total bilirubin in the body is available to exchange transfusion and that equilibration takes place at approximately 75 per cent of the original value. First, it is apparent that if the rate of rise in the level of serum bilirubin following an exchange transfusion is to be used as the guide to a second transfusion, the point of origin of this rate of rise must be taken to be approximately 75 per cent of the value in effect at the beginning of the first transfusion. Second, it is apparent that if a series of exchange transfusions are to be undertaken in an attempt to restore an established high level of serum bilirubin to safe levels, the number of normal exchange transfusions which will be necessary to restore a level of 30 mg. per 100 ml. to a sustained level below 20 mg. per 100 ml., *in the absence of further generation of bilirubin*, will be two exchange transfusions, whereas at least three exchange transfusions will be necessary to restore a

level of 40 mg. per 100 ml. to a sustained level of less than 20 mg. per 100 ml. Given the rapidity with which the equilibration of bilirubin occurs in the body and the further consideration that the largest amount of bilirubin is removed early in the course of an exchange transfusion when serum levels are highest, it will be appropriate in the control of an established high level to consider the need for serial transfusions at intervals of three to four hours and sometimes to divide a given pint of blood into two aliquots, which may be used at three- to four-hour intervals so as to take maximal advantage of the opportunity to remove bilirubin at a high serum level.

When exchange transfusions are done solely for the purpose of removal of bilirubin, whole blood is the preferred medium. There should be *no* need to leave a deficit in blood volume in such infants at the end of the transfusion; this is helpful only early in care of severely ill infants with elevated venous pressure. Indeed, a modest excess may sometimes be offered the infant to replace blood withdrawn for laboratory studies.

MANAGEMENT OF UNEXPECTED HEMOLYTIC DISEASE—A AND B INCOMPATIBILITY AND THE LIKE

Unexpected hemolytic disease usually comes to light when the newborn infant becomes icteric too soon or in a too conspicuous degree; only exceptionally will unexpected disease present obvious signs of a hemolytic process at the time of delivery. This will be most likely to occur when the usual variety of Rh incompatibility (that due to anti-Rh_0 or anti-D) has not been adequately anticipated through oversight or through laboratory error. Incompatibility with respect to A or B causes by far the largest part of unexpected hemolytic disease, but this is almost never conspicuous at birth. It has been amply demonstrated that hemolytic disease due to A or B incompatibility is, in fact, the most common clinical variety.

Halbrecht[12] first showed that the cardinal clinical feature of hemolytic disease due to A or B incompatibility was the appearance of icterus within the first 24 hours of life, and the presumption seems well justified that any infant with conspicuous icterus prior to the thirty-sixth hour of life, in whom Rh incompatibility is known not to exist, has A or B incompatibility until proved otherwise.

Case finding in A or B incompatibility resides in the systematic careful observation of newborn infants for the time of onset of or unusual intensity of icterus. This will generally be most effectively accomplished when a routine survey of the nursery for icteric infants is made with each change of nursing personnel. At Temple University Hospital, Vaughan and Brelian[13] found that 35 per cent of white infants and 15 per cent of Negro infants displayed clinical icterus within the first 6 days of life and that there was a preponderance of icteric male infants among the white group. In both groups, surprisingly, approximately 10 per cent of those infants who became icteric within the first 6 days of life were first observed to be jaundiced within the first 24 hours[11]; except where hemolytic disease was present this jaundice was quite mild in degree. On the other hand, we know two instances of hemolytic disease in which B incompatibility was responsible for kernicterus in infants in whom

icterus was not noted until the third day of life, in one of whom we can be quite certain that icterus was not present at 48 hours of age. Neither, therefore, does the early onset of icterus certify to the diagnosis of hemolytic disease nor the late onset exclude it.

Zuelzer and Cohen have summarized well the difficulties of substantiating a clinical impression of hemolytic disease due to A or B incompatibility; they have indicated those results of examination of maternal serum or of infant's blood that are likely to be helpful in diagnosis. The mother in such instances will usually prove to belong to group O, the infant to A or B; the mother will of course have anti-A and anti-B antibodies. It will be helpful if it can be shown that the mother has no other blood group antibody in her serum. The direct antiglobulin reaction (Coombs test) on the infant's cells is generally negative in A or B incompatibility unless special techniques or sera are used. The infant's cells often show marked spherocytosis and there may be conspicuous reticulocytosis. Osmotic fragility may be increased. The red blood cells of the infant may show spontaneous agglutinability in certain colloidal media such as adult serum, acacia, or glue.

Unfortunately, even when all of these examinations are carried out, except for the demonstration of incompatibility between mother and infant, the diagnosis of A or B incompatibility may rest on insecure grounds. On the other hand, the therapeutic problem may be quite urgent when the bilirubin level exceeds or threatens to exceed 20 mg. per 100 ml. Under these circumstances, it will generally seem appropriate to meet the threat of kernicterus, which is real, with exchange transfusion directed at the control of the level of bilirubin in exactly the same manner as described above for Rh incompatibility. This is appropriate until and unless it can be shown that the threat of kernicterus is different in this condition from what it is in Rh incompatibility at any given level of serum bilirubin or that some other procedure is more appropriate. The same considerations govern the choice of therapy for unexpected hemolytic disease which is due to blood group incompatibility other than A or B.

It is worthy of note that *whatever* the nature of the incompatibility which may be responsible for hemolytic disease in a newborn infant, it is *always* possible to find an appropriate donor for the exchange transfusion to be given to the infant, even when the exact nature of the blood group incompatibility is unknown and may not be discovered for some weeks. ***The blood that will be appropriate for the infant is one which is compatible with his mother's serum, by the indirect antiglobulin (Coombs) technique.***

HYPERBILIRUBINEMIA UNASSOCIATED WITH HEMOLYTIC DISEASE

In recent years increasing attention has been focused upon the possible threat of kernicterus to newborn infants who have hyperbilirubinemia which is not the result of hemolytic disease. In premature infants, particularly, kernicterus has attracted increased attention, and it has been convincingly demonstrated that kernicterus in premature infants may have been disposed to by certain routine practices which clearly ought to be

abandoned, such as the use of sulfisoxazole in prophylaxis or treatment of infection,[14] or the use of unnecessarily high doses of synthetic vitamin K in the prophylaxis of hemorrhagic disease.[15] It remains to be seen what other modifications of management of premature infants may be necessary to reduce the likelihood of hyperbilirubinemia and kernicterus to a minimum. Meanwhile, some anxiety attends the observation of high levels of bilirubin in premature infants in whom disposing factors may or may not exist. We have taken the position that a bilirubin level in excess of 20 mg. per 100 ml. is intolerable, without respect for its reason for existence, and have used exchange transfusion prophylactically against the possible development of kernicterus in infants who present such levels.

SOME PRACTICAL POINTS REGARDING EXCHANGE TRANSFUSION

Only in instances of urgent need should blood be used for the exchange transfusion of an infant with suspected hemolytic disease which has not been shown to be compatible with maternal serum by the indirect antiglobulin (Coombs) technique. Only through demonstration of this compatibility with maternal serum can the possibility be excluded of blood being used which is incompatible by virtue of the mother's multiple sensitization or her sensitization to a blood group antigen which is not suspected. The infant with Rh incompatibility will receive blood of group O if his mother is group O or if he himself is group O; on the other hand, if both he and his mother belong to group A, it may be found more satisfactory to use group A, Rh-negative blood. If both infant and mother belong to group B, group O, Rh-negative blood should be used for the infant unless it can be assured that sufficient group B, Rh-negative blood is available to meet any conceivable need for multiple exchange transfusions. We believe that we have seen heightened bilirubinemia following a second exchange transfusion using group O, Rh-negative blood in a group B infant whose first exchange transfusion used the last available pint of group B, Rh-negative blood.

We prefer to have the donor blood as fresh as possible and will generally reject blood which has been stored for more than four days, owing to the hyperkalemia which attends longer storage. Ideally, the suitability of a donor ought to be established prior to delivery and the blood drawn at the time of delivery so that it can be given to the infant at body temperature. Some means for warming chilled blood ought to be available; its partial immersion in a water bath at 37° C. *(never higher)* will generally suffice.

Prophylactic administration of antibiotic agents following exchange transfusions seems demanded by the nature of the procedure, especially after the first few hours of life. We have administered 50,000 U. of aqueous penicillin and 50 mg. of streptomycin every 12 hours unless information concerning the prevalent bacterial flora suggested that other agents ought to be used. Present evidence suggests that sulfonamide drugs should under no circumstances be used. The administration of prophylactic drugs ought to continue for 72 hours following the last exchange transfusion.

Multiple exchange transfusions will generally be most satisfactorily carried out by the umbilical vein, using the apparatus described by Allen and Diamond. We have used this route as late as the eighth day and for as many as seven exchange transfusions. Its use is facilitated by the avoidance of placement of suture material in the wall of the vein or elsewhere in the region following exchange transfusion. An eminently satisfactory dressing consists of one or two 2 by 2 inch gauze squares held in place over the umbilical stump by a wide (1 inch) strip of adhesive tape which extends around the infant's trunk from one costal vertebral angle to the other and supplies gentle pressure. We have not encountered any misadventure in the use of this simple dressing. When the umbilical vein is to be re-entered, it is generally easily found in the twelve o'clock position on the umbilical stump which has been cut off to the level of abdominal wall. Re-entry is facilitated by the soaking of this area for a few seconds in saline solution.

AFTERCARE OF INFANTS WITH HEMOLYTIC DISEASE

In infants with hemolytic disease in whom the first week of life has been passed without the development of kernicterus (which almost always occurs before the sixth day), the outlook for complete recovery is excellent so long as adequate attention is given the possible development of anemia. Infants who have received exchange transfusions and who have hemoglobin levels of 9 Gm. per cent or above may generally be sent home, with the confident expectation that they will need no further treatment. There is generally a gradual fall to levels between 7 and 8 Gm. per cent during the next four to six weeks, at the end of which time a reticulocytosis will indicate the imminent restoration of normal blood values. In infants who have not received treatment, there may be some risk of a fall in hemoglobin during the second or third week of life to more dangerous levels, and instances are known of fatal collapse at that time, apparently owing to anemia. It is appropriate, therefore, to follow the progress of the hemoglobin level closely in such infants. We have given small transfusions of packed cells at levels below 9 Gm. per cent within the first three weeks or 7 Gm. per cent after three weeks, unless a reticulocytosis indicated active blood formation on the part of the infant.

There is usually no need for supplemental iron or other supportive therapy in infants with hemolytic disease; the more violent the hemolytic process, the less likely any need ever for supplemental iron.

EMOTIONAL CARE, PROGNOSIS, AND GENETIC COUNSELING IN HEMOLYTIC DISEASE

An essential element in the care of the family threatened with, or deeply involved in, hemolytic disease is the recognition of and, where honestly possible, the allaying of anxiety surrounding any current or future pregnancy. Less emotional support comes from a recitation of statistical probabilities than from a calm facing of all possible eventualities with assurances that everything necessary or possible will be taken into consideration in planning for the care of mother and infant.

The prognosis of hemolytic disease due to A or B incompatibility is so good, with adequate treatment, as well

as so unlikely to become worse with subsequent pregnancies that there is no good reason to be other than optimistic as to the future, with the assurance that whatever bridges may need to be crossed are likely to be easily traversed.

With regard to Rh isoimmunization, the situation is more complex. If Rh antibody is not demonstrated in the Rh-negative woman, she may be assured that illness in the infant due to Rh incompatibility need be of no present concern and that in the unlikely event that the antibody should ever appear, appropriate plans will be made. Once the antibody becomes demonstrable, regardless of titer or history, a hopeful attitude is proper, but a simple pat-on-the-back reassurance that everything will be all right is definitely not.

The sensitized mother deserves to know that sometimes the condition of the infant becomes so serious before delivery that he is not born alive. She may otherwise remember with great bitterness the profound shock of the unexpected stillbirth of the infant. Such mothers often feel that this possibility, however unlikely, ought to have been called to their attention. Nothing more than casual mention of this as a possibility seems necessary, unless it seems to threaten in such degree as to raise concern as to whether some modification of management of pregnancy may be necessary. The sensitized mother who has had a previous stillbirth generally knows that however unhappy the immediate experience, it is tolerable in retrospect.

Every sensitized mother deserves to know that if her infant is born alive and in good condition his recovery is overwhelmingly likely. Many will have heard that recovered infants have cerebral palsy or a mental defect. They can be told that kernicterus is preventable almost always with modern treatment and that if treatment can be carried out according to usual plan, it is so unlikely that it need not enter seriously into present consideration. Such a declaration can be followed with the observation that "it's natural for you to worry just a little bit about that, just the same, isn't it?"

The outlook for future infants of the mother whose infant has recovered from, or has succumbed to, hemolytic disease due to Rh incompatibility can only be described as uncertain. A recovered infant offers a hopeful outlook, a fatality a less hopeful prognosis. Some families have many mildly affected infants, others only very severely affected ones. There is no family history, on the other hand, so encouraging that a fatality may not occur in another pregnancy; nor is there any family history so bad that an unexpected satisfactory result may not occur, possibly as a result of some modification of management. It seems better that uncommitted parents who understand these issues come to decisions as to future conduct which depend upon their *own* appraisals of their desires and strengths and tolerances rather than that the physician strongly urge one or another course of action which may better reflect his own concern or need than his patients'.

If the father of a potentially ill fetus can be shown to be heterozygous, the likelihood of an Rh-negative normal infant becomes 50 per cent. This should not be taken as meaning, however, that under this circumstance parents ought to be more freely encouraged to try further pregnancies once

the mother is sensitized. No assurance of satisfactory issue derives from this information; we have encountered five successive stillborn Rh-positive fetuses in the offspring of an Rh-positive man ultimately proved heterozygous by the delivery of a normal Rh-negative infant. Parents who cannot accept such results, whatever the statistical risk, should not participate in this kind of genetic game. Those who do elect further adventures in hemolytic disease deserve our best and most thoughtful help and support.

SUMMARY

The management of hemolytic disease of the newborn infant has been reviewed with respect to the three major problems which may be presented: the prevention of stillbirth, the management of the infant severely ill at birth, and the prevention of kernicterus in the infant with hyperbilirubinemia. Each of these circumstances demands its own special orientation to the infant's immediate need. Consideration is given the needs of involved parents for emotional support and counseling in family planning.

REFERENCES

1. Vaughan, V. C. III, Allen, F. H., Jr., and Diamond, L. K.: Erythroblastosis Fetalis. I. Problems in the Interpretation of Changing Mortality in Erythroblastosis Fetalis, Pediatrics **6**: 173, 1950.
2. Mollison, P. L., and Walker, W.: Controlled Trials of Treatment of Hemolytic Disease of the Newborn, Lancet **1**: 429, 1952.
3. Allen, F. H., Jr.: Induction of Labor in the Management of Erythroblastosis Fetalis, Quart. Rev. Pediat. **12**: 1, 1957.
4. Chown, B., and Bowman, W. D.: The Place of Early Delivery in the Prevention of Foetal Death From Erythroblastosis, Pediat. Clin. North America **5**: 279, 1958.
5. Chown, B.: Anaemias in the Newborn Other Than Haemolytic Disease, Pediat. Clin. North America **4**: 371, 1957.
6. Wheeler, W. E., and Ambuel, J. P.: The Efficient Use of Exchange Transfusions in the Treatment of Erythroblastosis, Pediat. Clin. North America **4**: 383, 1957.
7. Mollison, P. L., and Cutbush, M.: Haemolytic Disease of the Newborn; Criteria of Severity, Brit. M. J. **1**: 123, 1949.
8. Allen, F. H., Jr., and Diamond, L. K.: Erythroblastosis Fetalis, Including Exchange Transfusion Technique, Boston, 1957, Little, Brown & Co.
9. Zuelzer, W. W., and Cohen, F.: ABO Hemolytic Disease and Heterospecific Pregnancy, Pediat. Clin. North America **4**: 405, 1957.
10. Brown, A. K., Zuelzer, W. W., and Robinson, A. R.: Studies in Hyperbilirubinemia. II. Clearance of Bilirubin From Plasma and Extravascular Space in Newborn Infants During Exchange Transfusion, A. M. A. J. Dis. Child. **93**: 274, 1957.
11. Vaughan, V. C. III, and Brelian, F. B.: Unpublished Data.
12. Halbrecht, I.: Role of Hemoagglutinins Anti-A and Anti-B in Pathogenesis of Jaundice of the Newborn (Icterus Neonatorum Praecox), Am. J. Dis. Child. **68**: 248, 1944.
13. Vaughan, V. C. III, and Brelian, F. B.: The Jaundiced Neonate: Differential Diagnosis and Management, *in* A. M. A. Scientific Exhibits of 1957, New York, 1957, Grune & Stratton, p. 313.
14. Silverman, W. A., Andersen, D. H., Blanc, W. A., and Crozier, D. N.: A Difference in Mortality Rate and Incidence of Kernicterus Among Premature Infants Allotted to Two Prophylactic Antibacterial Regimens, Pediatrics **18**: 614, 1956.
15. Crosse, V. M., Meyer, T. C., and Gerrard, J. W.: Kernicterus and Prematurity, Arch. Dis. Childhood **30**: 501, 1955.

NONHEMOLYTIC ANEMIA OF THE NEWBORN INFANT: HEMORRHAGE OF FETAL BLOOD INTO THE MATERNAL CIRCULATION

H. H. Gunson, M.B., Ch.B.*
Winnipeg, Manitoba, Canada

THE fetus may lose blood in one of several ways before or during the birth process. If the infant is born with anemia, the cause may be obvious. Blood may have been lost because of abruptio placenta, placenta previa, or tearing, rupturing, or cutting of a normal umbilical cord or unsupported vessels such as in velamentous insertion of the cord.[1] Occasionally the fetus may bleed less openly from one of the above causes, and this gives rise to what Chown[2] calls "mistaken fetal blood loss." Under these circumstances the observed hemorrhage is considered to be maternal in origin, whereas it is really coming from the fetus.

Truly concealed fetal hemorrhage, with no external bleeding before or during parturition, occurs when fetal blood enters the maternal circulation. This recently proved syndrome has been referred to as "fetomaternal transfusion." The first suggestion that fetal hemorrhage into the maternal circulation could be the cause of neonatal anemia was made by Wiener.[3] This mechanism was also referred to by Wickster and Christian[1] as a cause of severe posthemorrhagic anemia in newly born infants. Laski and Johnston[4] reported twenty-two cases of nonhemolytic anemia in neonates in which a presumptive diagnosis of this syndrome was made.

Proof for the supposition that fetomaternal transfusion was indeed an entity was furnished by Chown[5] in studying an infant suffering from moderately severe anemia at birth. By means of differential agglutination and estimation of alkaline-resistant hemoglobin, he was able to prove conclusively the presence of fetal erythrocytes in the mother's blood. Subsequently, Chown[6] reported a case of neonatal anemia with the fetal vessels hemorrhaging primarily into the substance of the placenta, together with fetal red cells in the intervillous sinuses adjacent to the site of hemorrhage.

Since this work there have been several reports of transfusion. The majority of cases have been infants with moderate or severe anemia at the time of birth, namely, Gunson,[7] Goodhall and his co-workers,[8] Grant and Reeve,[9] Zuelzer,[10] Borum and associates,[11] and McGovern and his colleagues.[12] In addition Schiller[13] described a clear-cut case of fetal hemorrhage into the maternal circulation which occurred immediately before delivery. The baby was in a state of

From the Rh Laboratory and the Department of Pediatrics, The University of Manitoba.

*Present address: The Regional Transfusion Centre, Roby Street, Manchester 1, England.

shock but was not anemic. Dunsford[14] by means of delicate immunohematologic tests very neatly demonstrated the presence of fetal erythrocytes in the blood of four mothers whose infants were clinically normal. O'Connor and co-workers[15] and Colebatch, Pitt, and Maddison[16] have reported cases of mild or moderate anemia which they believed to be due to fetomaternal transfusion. Proof for their diagnoses depended on slight elevations of alkaline-resistant hemoglobin and must be regarded as equivocal.

There is little doubt that fetal red cells can reach the maternal circulation in considerable numbers. The mechanics of their passage has not been fully explained. Kline[17] examined the placentas of 227 infants of whom 15 suffered from erythroblastosis fetalis. He found breaks in the placental barrier in all the specimens and nucleated red cells in the intervillous spaces. This does not explain, however, how blood passes in the direction from fetus to mother. Perhaps this is controlled in some way by the intrauterine and intraplacental pressures developed by the gravid uterus at term and during labor. There is evidence, albeit equivocal in some instances, that blood can cross the placenta in the opposite direction, i.e., from mother to fetus. This aspect of transplacental passage of red cells has been investigated by injecting erythrocytes tagged with radioactive phosphorus (P^{32})[18] and radioactive iron (Fe^{59})[19] into the mother's blood just before delivery of the infant. An assessment of the passage of red cells from the maternal to the fetal circulation was made by the demonstration of radioactivity in the cord blood. These methods, particularly using P^{32}, have the disadvantage of dissociation of the radioactive material from the cell and lysis of the cells with subsequent liberation of radioactive material into the plasma. Another approach to this problem has been the injection of blood with the sickling trait[19] and elliptocytosis[20] into mothers and recovering the abnormal cells from the cord blood of the infant. To my knowledge no one has conclusively demonstrated the presence of maternal erythrocytes in the fetal circulation by differential agglutination.

CLINICAL MANIFESTATIONS

The clinical signs the infant exhibits naturally depend on the quantity and rate of the blood loss and the time interval elapsing after the hemorrhage and before the birth of the baby. Rapid hemorrhage from the fetus immediately before birth will lead to a state of shock, whereas prolonged or repeated hemorrhage while the infant is in utero produces a well-established anemia at the time of birth. In the majority of proved cases anemia rather than shock has been the presenting feature.

The anemia comes as a surprise to the attending physician since there is nothing more than normal blood loss noticeable during delivery of the infant. In more severely anemic infants there is pallor. The baby is inactive and resuscitation is slow. The cry is weak and breathing is rapid and shallow. The liver and spleen are not usually enlarged. One must be careful not to mistake the clinical picture at this stage for asphyxia pallida. If the hemorrhage has been less severe, the symptoms will be proportionately reduced. The infant may

seem normal at birth and then the pallor noticed after a few hours.

The clinical course of these infants is characteristic. Since blood has been lost from the fetal surface of the placenta, the simple procedure of tying off the cord vessels stops the bleeding. Therefore, the anemia is not progressive, and the tendency is for the hemoglobin to rise following birth because of the intense erythropoietic activity in the infant's tissues. The latter phenomenon can be demonstrated by examination of the blood smear which shows reticulocytosis and increased numbers of nucleated red cells.

DIAGNOSIS

Anemia in a newborn infant may be due to hemorrhage, hemolytic disease, or hypoplastic anemia. Differentiation between the various types of hemorrhage is not usually difficult. Fetomaternal transfusion is an unexpected concealed hemorrhage while other causes of bleeding arise from pathologic conditions which are recognizable before the infant is born or immediately after birth has occurred. The clinical picture of a severe anemia at birth closely mimics shock, and steps should be taken to determine the hemoglobin concentration of the baby since the prognosis and treatment of the two conditions are considerably different.

Hemolytic disease of the newborn infant due to Rh incompatibility may produce severe anemia. There is usually yellow staining of the cord and the amniotic fluid, and the liver and spleen are enlarged. The more severely affected infants may be jaundiced at birth and have purpura. A positive Coombs test is the best single procedure to differentiate this disease from posthemorrhagic anemia. It is unlikely that confusion of the two syndromes will occur except when the hemolytic disease is severe enough to cause death of the infant if treatment is not established immediately. ABO hemolytic disease does not usually present with anemia but is more commonly recognized as a mild early jaundice. A positive Coombs reaction, when this test is done in the capillary,[21] and the presence of homologous anti-A or anti-B in the serum of the infants,[22] together with microspherocytosis are characteristics of this type of erythroblastosis.

Hypoplastic anemia is uncommon in newborn infants but has been reported.[23, 24] The presence of regenerative erythrocytes in the peripheral smear rules out this condition, and their absence rules out anemia due to hemorrhage.

Thus, by taking a careful maternal history to rule out a situation favorable for fetal bleeding and a clinical examination of the infant, a presumptive diagnosis of anemia due to bleeding of the fetus into the maternal circulation can be made. Quite often this is all that can be done before the infant is treated, since absolute confirmation of the diagnosis is often time consuming. The proof that fetal cells have entered the maternal circulation has been approached, in the main, in two ways: by using the method of differential agglutination and by determination of an excess of alkaline-resistant or fetal type of hemoglobin.

The method of differential agglutination is the one of choice if conditions are suitable for its use. An

obvious prerequisite is that there is a major blood group difference between the mother and her infant. Thus, if the mother is group A, Rh negative and her infant is group O, Rh positive (probable genotype cDE/cde), as in the case I[7] reported, exposure of the mother's red cells to anti-D and anti-E is sufficient to demonstrate the fetal antigens in the maternal blood. Moreover, in this case, by adding anti-A to the mother's erythrocytes, the fetal group O red cells could be separated and their number estimated. These are, of course, ideal circumstances, but many combinations of groups of the mother's and the infant's blood will allow the use of this method. Limitations even in the presence of antigenic differences may be encountered where a mother has an antibody antagonistic to the infant's red cells. Thus, if the mother is group O and the infant is group A or B, the maternal isoantibodies are theoretically capable of destroying the fetal cells. One would have to be lucky enough to carry out the investigations shortly after the hemorrhage had occurred. Under these circumstances, Dunsford's[14] technique for demonstrating the presence of fetal erythrocytes in the maternal circulation may well be very useful. He found that a group O mother who had delivered a group A infant lacked the expected anti-A agglutinins immediately after the baby was born. He was able to substantiate his assumption from this finding, that fetal red cells were in the mother's blood, by direct differential agglutination. Ten days post partum the so-called immune type of anti-A was present in the mother's serum. Subsequently, Dunsford found three other cases where the isoantibody in the mother's serum corresponding to the infant's ABO group was absent immediately following parturition. In none of the cases was the baby anemic. A different type of approach was made by Goodhall and associates[8] who found Coombs positive red cells in the mother's blood in a case of Rh isoimmunization and erythroblastosis. He also demonstrated phagocytosis of nucleated red cells in the buffy coat of the mother's blood. Since she was not anemic, there was good reason to believe that these red cells were fetal in origin.

The use of the method of alkaline resistance of fetal blood (Singer and his colleagues[23]) may give definite results if there has been sufficient bleeding into the mother. This method has the disadvantage that most adults have up to 2 per cent of hemoglobin which is resistant to treatment with strong alkali, and values well in excess of this must be found before one can be sure that the increase is due to the presence of fetal blood in the adult circulation. It is, however, a method which can be used independently of blood groups and should be tried whenever the diagnosis is in doubt and also as a confirmatory method for the differential agglutination test.

Another possibility which has to be remembered is that the hemorrhage may not have entered the general circulation of the mother, as in the case described by Chown[6] where hemorrhage into the placenta occurred. Thus, in all cases where the newborn infant is anemic, the placenta should be given more than the usual cursory

examination by a person who is familiar with the complexities of this organ.

TREATMENT

The treatment of the infant depends on the degree of anemia. Many infants require no specific treatment, and blood transfusion where the hemoglobin concentration is in the range of 8 or 9 Gm. per cent may serve to depress marrow function. The more severely anemic infants will require transfusion, and in those with very low hemoglobin concentrations associated with a high output heart failure, exchange transfusion is advisable. During this procedure, the venous pressure can be lowered at the same time the anemia is corrected. This method of treatment is often lifesaving and cannot be too highly recommended.

When faced with a severely anemic infant, it is difficult not to run to the emergency blood bank and grasp a bottle of group O, Rh negative blood and start the transfusion. It is much preferable to give group compatible blood to these infants. In the time it takes to set up the intravenous infusion ABO grouping can be done. If one gives group O blood to a group A or B infant, there is a risk of the infant developing jaundice due to the minor incompatibility. This can occur even if the isoantibody titer of the donor blood is low. Fortunately, the jaundice is not usually severe and the infants escape serious consequences; nevertheless, this is a form of iatrogenic ABO hemolytic disease which causes an abnormally rapid destruction of the infant's cells—a state of affairs which is undesirable, to say the least. If there are no facilities for rapid grouping of the infant's blood, the mother's blood can be used. This will always be compatible with the infant's blood but has the same disadvantages as group O, Rh negative blood, in that the mother may be group O and the infant group A or B. Moreover, under these circumstances the mother may have a high titer of isoagglutinins.

COMPLICATIONS

Maternal Isoimmunization.—In 1941 Levine and his co-workers[26] proposed an etiology for hemolytic disease of the newborn infant which depended on the entry of fetal erythrocytes into the maternal circulation. In two of the published cases of anemia due to fetomaternal transfusion, formation of Rh antibodies by the mother has been a sequela.[5, 7] In Chown's case[5] they appeared in the mother's serum at 22 days post partum, and in my own[7] at 80 days after the baby was born. Dunsford[14] reported the formation of immune anti-A subsequent to finding fetal red cells in the maternal circulation.

If erythrocytes from the fetus can pass into the maternal circulation in quantities sufficient to cause anemia in the infant, there is a distinct possibility that they may enter the mother's blood in much fewer numbers without causing clinical disease in the infant, yet still being sufficiently numerous to induce antibody formation in the mother. Creger and Steele[27] using a differential agglutination method in the combination of a group A mother and a group O baby found fetal cells in the maternal blood with surprising frequency. Hosoi[28] found evidence of fetal hemoglobin in the maternal circulation in

18 out of 47 mothers, using an immunological method for detecting the fetal hemoglobin. Both of these investigations suggest that fetal cells appear in the maternal circulation in many more instances than Rh antibodies, the latter phenomenon occurring in about 5 per cent of Rh negative mothers who carry Rh positive fetuses. The development of antibodies may depend on the number of fetal red cells gaining access to the maternal circulation and also the inborn ability of certain persons to produce antibodies following a specific stimulation.

Transfusion Reaction in the Mother.—In one probable case described by Chown[6] and a proved case of Goodhall's,[8] the mother had a febrile reaction with a rigor immediately before the birth of the infant. Both of these mothers had Rh antibodies in their serum and their infants were Rh positive. The assumption was that the febrile reaction was caused by the entry of incompatible red cells from the fetus into the maternal circulation, giving, in effect, a transfusion reaction. This is a reasonable interpretation and makes for a very interesting complication of fetomaternal transfusion.

Iron-deficiency Anemia.—McGovern and co-workers[12] reported the occurrence of iron-deficiency anemia in a 3-month-old infant who suffered anemia at birth from hemorrhage into the maternal circulation. The baby was transfused on the first day of life, and the iron-deficiency state was also treated initially by transfusion. Since his hemoglobin concentration was 7 Gm. per cent when this was diagnosed, perhaps a more logical form of treatment would have been to give the baby iron instead of blood.

SUMMARY

Nonhemolytic anemia of the newborn infant due to fetal hemorrhage into the maternal circulation is a real clinical entity. It may be much commoner than the paucity of reports leads one to think. Every effort should be made to establish the correct diagnosis in the severely anemic infants an soon after birth as possible, since this condition is eminently curable. Perhaps it is not too unfortunate that the mild cases are sometimes missed, because the self-limiting nature of the disease will cure the infant without the help of vigorous and often unnecessary transfusions.

REFERENCES

1. Wickster, G. Z., and Christian, J. R.: Post-hemorrhagic Shock in the Newborn Period, Pediat. Clin. North America **1**: 555, 1954.
2. Chown, B.: Anemias in the Newborn Other Than Haemolytic Disease, Pediat. Clin. North America, May, 1957, p. 371.
3. Wiener, A. S.: Diagnosis and Treatment of Anemia of the Newborn Caused by Occult Placental Hemorrhage, Am. J. Obst. & Gynec. **56**: 717, 1948.
4. Laski, B., and Johnson, W. H.: Twenty-two Cases of Nonhemolytic Neonatal Anemia, A. M. A. Am. J. Dis. Child. **88**: 811, 1954.
5. Chown, B.: Anemia From Bleeding of the Fetus Into the Mother's Circulation, Lancet **1**: 1213, 1954.
6. Chown, B.: The Fetus Can Bleed: Three Clinical Pictures, Am. J. Obst. & Gynec. **70**: 1298, 1955.
7. Gunson, H. H.: Neonatal Anemia Due to Fetal Hemorrhage Into the Maternal Circulation, Pediatrics **20**: 3, 1957.
8. Goodhall, H. B., Graham, F. S., Millar, M. C., and Cameron, C.: Transplacental Bleeding From the Fetus, J. Clin. Path. **11**: 251, 1958.
9. Grant, R. T., and Reeve, E. B.: Observations on the General Effects of Injury in Man, Med. Res. Council. Sp. Rep. Ser. 227, 1951.
10. Zuelzer, W. W.: Quoted by Chown.[2]

11. Borum, A., Lloyd, H. O., and Talbot, R. T.: Possible Fetal Hemorrhage Into the Maternal Circulation: Report of Two Cases, J. A. M. A. **164**: 1087, 1957.
12. McGovern, J. J., Driscoll, R., Dutoit, C. H., Grove-Rasmussen, M., and Bedell, R. F.: Iron Deficiency Anemia Resulting From Fetomaternal Transfusion, New England J. Med. **258**: 1149, 1958.
13. Schiller, J. G.: Shock in the Newborn Caused by Transplacental Hemorrhage, Pediatrics **20**: 7, 1957.
14. Dunsford, I.: Proof of Fetal Antigens Entering the Maternal Circulation, Vox sanguinis **2**: 125, 1957.
15. O'Connor, W., Sheilds, G., Schuyler, K., and Sussman, K.: The Occurrence of Anemia of the Newborn in Association With the Appearance of Fetal Hemoglobin in the Maternal Circulation (Abst.), A. M. A. J. Dis. Child. **93**: 10, 1957.
16. Colebatch, J. H., Pitt, D., and Maddison, T. G.: Fetomaternal Transfusion: Another Cause of Post-hemorrhagic Anemia in the Newborn, M. J. Australia **2**: 209, 1956.
17. Kline, B. S.: Microscopic Observations of the Placental Barrier in Transplacental Erythrocytotoxi Anemia (Erythroblastosis Fetalis) and in Normal Pregnancy, Am. J. Obst. & Gynec. **56**: 226, 1948.
18. Naeslund, J., and Nylin, G.: Investigations on Permeability of Placenta With the Aid of Red Blood Corpuscles Tagged With Radio-active Phosphorus, Acta med. scandinav. Suppl. 170, p. 390, 1946.
19. Mengert, W. F., Rights, C. S., Bates, C. R., Reid, A. F., Wolf, G. R., and Nabors, G. C.: Placental Transmission of Erythrocytes, Am. J. Obst. & Gynec. **69**: 678, 1955.
20. Hedenstedt, S., and Naeslund, J.: Investigation of the Permeability of the Placenta With the Help of Elliptocytes, Acta med. scandinav. Suppl. 170, p. 126, 1946.
21. Chown, B., and Lewis, M.: The Slanted Capillary Method of Rhesus Blood Grouping, J. Clin. Path. **4**: 464, 1951.
22. Gunson, H. H.: An Evaluation of the Immunohematological Tests Used in the Diagnosis of AB Hemolytic Disease, A. M. A. J. Dis. Child. **94**: 123, 1957.
23. Smith, C. H.: Hypoplastic and Aplastic Anemias of Infancy and Childhood: With a Consideration of Non-hemolytic Anemia of the Newborn, J. PEDIAT. **43**: 457, 1953.
24. Kohlbry, C. O.: Congenital Hypoplastic Anemia, J. PEDIAT. **19**: 662, 1941.
25. Singer, K., Chernoff, A. L., and Singer, L.: Studies on Abnormal Hemoglobins; Their Demonstration in Sickle Cell Anemia and Other Hematologic Disorders by Means of Alkali Denaturation, Blood **6**: 413, 1951.
26. Levine, P., Katzin, E. M., and Burnham, L.: Isoimmunization in Pregnancy: Its Possible Bearing on the Etiology of Erythroblastosis Fetalis, J. A. M. A. **116**: 825, 1941.
27. Creger, W. P., and Steele, M. R.: Human Fetomaternal Passage of Erythrocytes, New England J. Med. **256**: 158, 1957.
28. Hosoi, T.: Serological Identification of Fetal Blood in the Maternal Circulation, Yokohama Med. Bull. **9**: 61, 1958.

PURE RED-CELL ANEMIA

CARL H. SMITH, M.D.
NEW YORK, N. Y.

AN UNUSUAL type of anemia is encountered in the first months of life which cannot be attributed to excessive blood destruction or blood loss but is classified with the hypoplastic-aplastic group by its refractory response to any treatment except transfusion of blood. Bone marrow examination reveals that the anemia is the result of a selective hypoplasia of red rell precursors. This circumstance in which an essential cellular element of the bone marrow is missing from birth lends itself to the same considerations with regard to causative teratogenic agents as other major congenital deformities.

Pure red-cell anemia,[20] one of the terms describing this entity,* refers to a chronic progressive anemia confined to a failure of erythropoiesis without an equivalent depression of the white blood cells or platelets.[7, 18, 33] The bone marrow is characterized by a complete or almost complete absence of cells of the erythroid series and normally proliferating cells of the granulocytic series and megakaryocytes. The failure of the bone marrow to provide adequate numbers of erythrocytes results in a persistent and progressive anemia with the need for repeated transfusions for a long time. Pure red-cell anemia has been observed at all ages, but it is particularly conspicuous as a congenital disease in early infancy and childhood.

From the Department of Pediatrics, The New York Hospital—Cornell Medical Center.

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*Other designations include chronic congenital aregenerative anemia, congenital hypoplastic anemia, pure red-cell aplasia, primary erythroid hypoplasia, and erythrogenesis imperfecta.

PATHOGENESIS

The etiology is unknown, but several theories have been offered to explain a condition in which but a single element of the bone marrow is affected. It is conceivable that the hematopoietic defect results from a noxious factor operating at critical periods of fetal development. In one of our cases, pure red-cell anemia occurred in conjunction with a hypoplastic and cystic kidney.[29] Presumably, the same factor that caused maldevelopment in the urinary tract was associated with the defect in erythropoiesis. In another group,[28] isoimmunization of a group O, Rh positive mother by an A, Rh positive offspring resulted in erythroblastosis. The maximum maternal anti-A agglutinin titer was 1:128,000. The subsequent course was typically that of pure red-cell anemia from which the child eventually died at 10 years of age. In this case at least, an antibody specifically directed against the red cells in fetal life may have caused prolonged and permanent depression of erythropoiesis. These circumstances are acknowledgedly unique but suggest a possible mechanism for the pathogenesis of the anemia.

Prolonged depression of erythropoiesis following Rh incompatibility in the newborn infant has been cited as an example of the persistent effect of antibody against erythrocyte precursors.[14] A rise in the reticulocyte count was associated with a significant drop in antibody titer. Other evidence of red-cell antibodies depressing erythrocyte precursors is the cases of complete[8] and temporary[13] pure red-cell anemia reported in association with autoimmune hemolytic disease. A temporary aregenerative crisis of one week's duration which results in severe anemia has been described in children in connection with toxic, infectious, or allergic etiologies.[11]

In the majority of cases, pure red-cell anemia which occurs in infancy and childhood may be compared with other congenital anomalies due to developmental defects. The most vulnerable period for damage to erythropoiesis is from the sixth to the twelfth week of fetal life, at which time the liver, spleen, and bone marrow are simultaneously involved in the proliferation of erythroblasts.[2, 15] Presumably, a noxious agent acting at this critical period selectively injures the red blood cell precursor.

Anthranilic acid, a metabolite formed in the breakdown of tryptophan, is excreted in relatively large quantities when the riboflavin-deficient animal is fed tryptophan. A similar defect in tryptophan metabolism has been reported in a number of cases of congenital pure red-cell anemia.[1] The administration of riboflavin to these patients resulted in a decrease in the excretion of anthranilic acid but did not alter the hematologic status.

CLINICAL FEATURES

The disease has an insidious onset with progressive pallor, anemia, irritability, listlessness, and anorexia which are usually apparent at the age of 2 to 3 months or later in the first year. The infant appears healthy at birth, and general development proceeds normally. Exceptionally, cases have been reported with a retarded onset, such as in a child 6 years old[16] and in two brothers at 17 years of age.[21] This occurrence in siblings has also been noted[3] in other cases.

Except for pallor, there is little to be observed on physical examination. The liver, spleen, and lymph nodes are not enlarged, jaundice is absent, and there are no bleeding manifestations. In some patients a particular type of facies has been described,[4] consisting of tow-colored hair, snub noses, thick upper lips, rather wide-set eyes, and an intelligent expression.

The liver and spleen enlarge as transfusion hemosiderosis develops. Normal red cell survival which is present when the spleen is not enlarged is shortened as this organ becomes the site of increased red cell destruction. The effect of long-standing administration of blood is to increase skin pigmentation. This is due both to iron deposition and to excessive melanin formation.[19] Cardiac enlargement, multiple arrhythmias, and refractory heart failure are inevitable consequences in many cases. While there is no definitive correlation between the development of cardiac complications and the amount of tissue iron resulting from transfusion therapy, it seems likely that both the persistent anemia and hemosiderosis contribute to the etiology The iron stores

accumulating from degraded hemoglobin are supplemented by increased iron absorption which is known to occur in hemolytic and aplastic anemias.

Pure red-cell anemia also occurs as an acquired erythrocytic hypoplasia in adults without a proved common etiology.[26, 31] Infection, allergy, red-cell autoantibodies in hemolytic anemia and chemical exposure have been incriminated. A more constant finding is the coexistence of this disease with benign thymoma.[5, 17, 25]

LABORATORY FINDINGS

The cardinal feature is a chronic aregenerative and refractory anemia in which there is a striking absence of nucleated precursors of the red cells from the bone marrow without a simultaneous depression of granulocytes or platelets or of their precursors. The anemia is normocytic and normochromic with low or absent reticulocytes. Without supportive transfusions, the hemoglobin level falls to extremely low values.

The number of platelets and white blood cells and the differential counts are normal. Marked leukopenia and moderate depression of platelets occur in rare instances in which the spleen enlarges and manifests a hypersplenic function. The bone marrow in such a case shows normal myeloid activity and a normal number of megakaryocytes. In a 7½-year-old boy under observation, splenectomy resulted in a return to normal of leukocytes and platelets but erythropoiesis remained hypoplastic. The serum iron level is elevated with complete saturation of the latent iron-binding capacity as a result of prolonged transfusion therapy.

In this condition, also, the bone marrow frequently contains varying numbers of primitive cells termed hematogones.[27, 32] Usually smaller than the ordinary lymphocytes, they resemble them closely in size and morphology. They possess a dense homogeneous matlike nucleus and a narrow rim of nongranular basophilic cytoplasm which is usually entirely absent in lymphocytes. These primordial cells possibly represent progenitors of the erythrocytic series. In one instance following the use of cortisone, they were replaced by small numbers of normoblasts.

PATHOLOGIC FINDINGS

The essential feature is widespread hemosiderosis, at times associated with evidence of hemochromatosis. In one 10-year-old patient who died of heart failure, postmortem examination revealed the typical syndrome of hemochromatosis, including diabetes mellitus and skin pigmentation. The heart was greatly enlarged; many of the myocardial fibers contained iron pigment; there was scanty interstitial fibrosis; and occasionally necrosis of isolated fibers was present. In another 9-year-old child whose terminal illness was severe hepatitis, postmortem examination revealed massive liver necrosis and generalized hemosiderosis with fibrosis of the liver, pancreas, and thyroid. The myocardial fibers were hypertrophied, vacuolated, and contained moderate amounts of hemosiderin.

DIAGNOSIS

In a typical congenital case, the disease is fully established in early infancy. The need for frequent transfusions is soon found to be based on erythropoietic hypoplasia in the bone marrow. It may occasionally follow

erythroblastosis and be mistaken for the protracted depression of erythropoiesis which may accompany this disease. Pure red-cell anemia can be differentiated from the hemolytic anemias such as sickle-cell disease, thalassemia, and hereditary spherocytosis by the lack of splenomegaly and the absence of icterus, reticulocytes, and of morphologic abnormalities in the red cell.

There is as yet no unanimity as to the degree of erythroid hypoplasia which this anemia implies. While the designation of pure red-cell anemia relates to conditions in which the bone marrow is practically devoid of normoblasts (less than 2 per cent), in some reports these elements though reduced are nevertheless present in moderate numbers. The myeloid-erythroid ratio which should be necessarily high in a case may be only moderately reduced. The problem of precise designation is exemplified by one report[4] describing 5 children with congenital hypoplastic anemia under the heading of "erythrogenesis imperfecta" in which several bone marrow examinations showed a variation between erythroid hypoplasia and hyperplasia. In another study,[3] the cases are described under the heading of hypoplastic anemia, with normoblasts ranging from 1.2 to 13.8 per cent and with an arrest of maturation at the polychromatic stage. These cases, however, share the common feature of varying aplasia of erythropoiesis without involvement of leukocytes or platelets.

Aplastic anemia is characterized by a simultaneous depression of the three principal cellular elements in the bone marrow; this results in a peripheral picture of profound anemia, leukopenia, neutropenia, and thrombocytopenia. Hypoplastic anemia differs from aplastic anemia in that the formation of red cells is impaired with lesser involvement of granulocytes and platelets. This term has been applied in recent years to intermediate conditions in which the three blood elements of the bone marrow are involved in variable degree. Reports of cases of hypoplastic anemia now range from those limited to a failure of red cell production to those in which leukocytes and platelets are simultaneously depressed but to a lesser degree than the red cells. In a restrictive terminology, pure red-cell anemia must be regarded as an aregenerative anemia in which the depression is confined exclusively to red cell production.

TREATMENT

Major therapy consists of transfusions, ACTH and steroids, and splenectomy.

Transfusions.—Supportive transfusions to maintain the hemoglobin at levels compatible with freedom from symptoms constitute a basic need. The use of sedimented or packed cells is preferable to whole blood. Although physiologic requirements for the growing child imply a need for optimum hemoglobin levels, this objective cannot be achieved without the danger of "iron overload" or hemosiderosis. The hemoglobin value at which a transfusion is indicated cannot be arbitrarily fixed but varies with the associated signs and symptoms of the individual patient. As a guide to management we found that with few exceptions patients in this category of chronic anemia do not require transfusions until hemoglobin levels drop to 7 to 7.5 Gm./100 ml., at which point clinical

symptoms usually appear. These include anorexia, listlessness, apathy, and incipient signs of heart failure. It is apparent that hemosiderosis is to be expected from repeated transfusions. Another limitation of frequent transfusions is their potential depressant effect upon endogenous erythropoiesis and hemoglobin synthesis.[31]

ACTH and Steroids.—In association with transfusions, adrenocortical hormones and ACTH are given primary consideration since they represent a potent form of therapy.[12, 24] Depending on the age, the steroid hormones (prednisone, prednisolone, and like synthetic substances) are prescribed in a daily dosage of from 15 to 60 mg., and are given in divided doses every 6 hours (a total of from 15 to 20 mg. in children under 2 years of age, and up to 60 mg. in older children). With optimal dosages, evidences of the ability of the bone marrow to produce red cells should be manifest within 3 to 4 weeks. If and when the patient responds, the dosage should be reduced to the minimum at which a remission is maintained—approximately 15 mg. daily in divided doses.

Under present trial are new synthetic corticosteroids as substitutes for prednisone (Meticorten) and prednisolone (Meticortelone). These are triamcinolone, Aristocort (Lederle), and Kenacort (Squibb); methylprednisolone, Medrol (Upjohn), and dexamethasone, Decadron (Merck Sharp & Dohme). It is claimed that the newer compounds require smaller dosage and produce a lesser incidence of side effects. In our experience, thus far, no superiority has been demonstrated over the older corticosteroids in the treatment of blood disorders in children such as thrombocytopenic purpura and leukemia. The 4 mg. tablet of Aristocort, Kenacort, and Medrol corresponds to the 5 mg. tablet of prednisone so that if trial is contemplated, the dosage should be adjusted on the basis of four-fifths of Meticorten. One 0.75 mg. tablet of Decadron replaces 4 mg. of methylprednisolone or triamcinolone or 5 mg. of prednisone or prednisolone.

A favorable response is reflected in a bone marrow and blood remission: erythroid hyperplasia, reticulocytosis, increased hemoglobin, and red cell levels. Any evidences of infection should prompt the administration of a suitable antibiotic. Despite the persistently low hemoglobin levels, these patients are not unduly susceptible to infection. If no response is obtained with the steroids within the prescribed period, ACTH by intramuscular or subcutaneous injection should be substituted, preferably as the gel in daily dosage of 40 mg. Here, too, the response varies with each patient so that no fixed amount can always be prescribed. With a favorable response, reducing and stopping the drug entirely should be attempted periodically to determine the capacity of the patient to establish spontaneous erythropoietic function.

It has been recommended that steroids be given early in the course since no response is to be expected when hemosiderosis has resulted after prolonged transfusion therapy.[13] Of 6 cases in our experience with congenital pure red-cell anemia, only one responded favorably to steroid therapy, and this occurred in a 17-month-old patient. In another case of this group of equal age, this treatment was ineffective. In the remaining 4 patients,

steroids were given at various intervals during the course without effect.

Splenectomy.—This procedure which is frequently recommended only as a last resort in pure red-cell anemia has nevertheless proved effective on occasion in both reducing the number of transfusions and in restoring erythropoiesis.[8, 21] When transfusions are required at increasingly frequent intervals to maintain a physiologically sufficient although reduced hemoglobin level, it is postulated that an extracorpuscular hemolytic component has developed which is presumably located in the spleen. Accelerated destruction can be confirmed by tagging normal donor red cells with radioactive chromium and noting their shortened survival.

In one of our patients with pure red-cell anemia, marked enlargement of the spleen was present. This finding, the failure to palpate the spleen, is contrary to an accepted criterion of the aplastic-hypoplastic group of anemias. In this instance, however, removal of the spleen resulted in extending the interval between transfusions, although depression of erythropoiesis persisted. Undoubtedly in cases without enlargement, splenectomy is justified where a hemolytic factor for donor blood is demonstrated by techniques which reveal a shortened survival of transfused normal erythrocytes. The presence of such a hemolytic factor would be clinically suspected when regularly spaced intervals for transfusions are suddenly shortened.

For patients in whom no hemolytic component is apparent, splenectomy is based on the principle that an abnormally functioning spleen may depress the erythropoietic function of bone marrow. This procedure is recommended when ACTH, the steroids, and other therapeutic agents alone or in combination have received extensive trial without success. There is less reluctance to remove the spleen when the bone marrow is not entirely devoid of erythroid elements in any stage of development. In any event, the procedure may be justified on the basis of a splenic humoral factor which inhibits maturation of precursors either visualized or potentially present in unidentified primordial cells such as hematogones. The fear that removal of the spleen deprives the patient of needed resources of erythropoietic tissue is not justified by experience in this disease. Although the majority of cases are not improved by splenectomy, the condition is not aggravated. Occasionally splenectomy alone has a beneficial effect in restoring erythropoiesis. In other cases, cortisone and other therapeutic agents have proved more effective after splenectomy. Increased experience may show that splenectomy will be efficacious in patients who respond initially to steroid therapy.[21]

These three agents constitute the mainstay of the therapeutic program in current use. Their skillful application has resulted in dramatic responses so that they have been useful in tiding the patient over until the time of a spontaneous and much-hoped-for remission.

Miscellaneous Treatment.—The agents cited by no means exhaust the entire therapeutic armamentarium of the disease. Worthy of note is the use of cobalt and members of the vitamin B complex. The administration of iron is potentially harmful because of

its increased absorption by the gastrointestinal canal in refractory anemias in which transfusions are supplementing the already excessive iron stores within the tissues.

The use of cobalt has had many advocates, with sporadic favorable reports in this and other chronic anemias. Its possible side effects such as anorexia, nausea, and thyroid enlargement preclude its widespread use except under controlled conditions.[23] Favorable results with cobalt, however, have been reported in several adults with the acquired form of this disease.[9, 26] In our experience, the results with cobalt in congenital pure red-cell anemia have been uniformly disappointing.

Each of the following vitamin B factors in the suggested dosage has been given to patients with this and other examples of the hypoplastic-aplastic group of anemias: B_{12} = 1,000 meg. intramuscularly 3 times weekly; riboflavin = 10 mg. 3 times daily[10]; nicotinic acid = 25 mg. once daily; and folic acid = 5 mg. 3 times daily. While many of these substances are known to function in specific phases of erythropoiesis, their value in this anemia is still debatable. Occasionally they appear to stimulate red cell formation, particularly after splenectomy.

PROGNOSIS

In most cases, the course of congenital pure red-cell anemia is one of chronic and progressive anemia with a fatal outcome. With supportive transfusion therapy, ACTH and steroids, and splenectomy, in selected cases, the patient can be maintained at a concentration of hemoglobin compatible with health. Long-term transfusions, however, carry the hazard of hemosiderosis and hemochromatosis. Reported cases suggest that a more favorable response to the steroid could be expected in cases with moderate numbers of normoblasts in the bone marrow than in those whose bone marrow was practically depleted of these cells. In one of our cases, however, no response with corticosteroids was obtained despite an initial finding of 5 per cent normoblasts.

An important factor in evaluating therapy and in a consideration of prognosis is the possibility of spontaneous recovery. Such remissions occurred in 3 of 12 patients cited by Diamond[6] and in other reports such as that of Palmen and Vahlquist.[22] Remission can occur at any period from early childhood to adolescence, with stabilization at a lower hemoglobin level.

Death results from cardiac failure, from hepatitis during the course of transfusions, and from overwhelming infection and sepsis. In these terminal events, the factor of hemosiderosis from excessive iron deposition resulting from transfusions and hemochromatosis may conceivably play an important complicating role.

REFERENCES

1. Altman, K. I., and Miller, G.: A Disturbance of Tryptophan Metabolism in Congenital Hypoplastic Anemia, Nature **172:** 868, 1953.
2. Arey, L. B.: Developmental Anatomy, ed. 4, Philadelphia, 1940, W. B. Saunders Co.
3. Burgert, E. O., Jr., Kennedy, R. L. J., and Pease, G. L.: Congenital Hypoplastic Anemia, Pediatrics **13:** 218, 1954.
4. Cathie, I. A. B.: Erythrogenesis Imperfecta, Arch. Dis. Childhood **25:** 313, 1950.
5. Clarkson, B., and Prockop, D. J.: Aregenerative Anemia Associated With Benign Thymoma, New England J. Med. **259:** 253, 1958.

6. Diamond, L. K.: Year Book in Pediatrics, 1954 to 1955 series, Chicago, Year Book Publishers, p. 308.
7. Diamond, L. K., and Blackfan, K. D.: Hypoplastic Anemia, Am. J. Dis. Child. **56**: 464, 1938.
8. Eisenmann, G., and Dameshek, W.: Splenectomy for "Pure Red-cell" Hypoplastic (Aregenerative) Anemia Associated With Autoimmune Hemolytic Disease, New England J. Med. **251**: 1044, 1954.
9. Fountain, J. R., and Dales, M.: Pure Red-cell Aplasia Successfully Treated With Cobalt, Lancet **1**: 541, 1955.
10. Foy, H., and Kondi, A.: A Case of True Red-cell Aplastic Anaemia Successfully Treated With Riboflavin, J. Path. & Bact. **65**: 559, 1953.
11. Gasser, C.: Akute Erythroblastopenie, 10 Fälle aplastischer Erythroblastenkrisen mit Riesenproerythroblasten bei allergisch-toxischen Zustandsbildern, Helvet. paediat. acta **4**: 107, 1949.
12. Gasser, C.: Aplastische Anämie (chronische Erythroblastophthise) und Cortison, Schweiz. med. Wchnschr. **81**: 1241, 1951.
13. Gasser, C.: Aplasia of Erythropoiesis. Acute and Chronic Erythroblastopenias or Pure (Red-cell) Aplastic Anaemias in Childhood, Pediat. Clin. North America, May, 1957, p. 445.
14. Giblett, E. R., Varela, J. E., and Finch, C. A.: Damage of the Bone Marrow Due to Rh Antibody, Pediatrics **17**: 37, 1956.
15. Gilmour, J. R.: Normal Haemopoiesis in Intra-uterine and Neonatal Life, J. Path. & Bact. **52**: 25, 1941.
16. Hansen, H. G.: Ueber die essentielle Erythroblastopenie, Acta haemat. **6**: 335, 1951.
17. Humphreys, G. H. II, and Southworth, H.: Aplastic Anemia Terminated by Removal of Mediastinal Tumor, Am. J. M. Sc. **210**: 501, 1945.
18. Josephs, H. W.: Anaemia of Infancy and Early Childhood, Medicine **15**: 307, 1936.
19. Lerner, A. B.: Melanin Pigmentation, Am. J. Med. **19**: 902, 1955.
20. Lescher, F. G., and Hubble, D.: A Correlation of Certain Blood Diseases on the Hypothesis of Bone Marrow Deficiency or Hypoplasia, Quart. J. Med. **1**: 425, 1932.
21. Loeb, V., Jr., Moore, C. V., and Dubach, R.: The Physiologic Evaluation and Management of Chronic Bone Marrow Failure, Am. J. Med. **15**: 499, 1953.
22. Palmen, K., and Vahlquist, B.: Stationary Hypoplastic Anemia, Acta haemat. **4**: 273, 1949.
23. Panels in Therapy: V. The Use of Cobalt and Cobalt-Iron Preparations in the Therapy of Anemia, Blood **10**: 852, 1955.
24. Pearson, H. A., and Cone, T. E., Jr.: Congenital Hypoplastic Anemia, Pediatrics **19**: 192, 1957.
25. Ross, J. F., Finch, S. C., Street, R. B., Jr., and Strieder, J. M.: Simultaneous Occurrence of Benign Thymoma and Refractory Anemia, Blood **9**: 935, 1954.
26. Seaman, A. J., and Koler, R. D.: Acquired Erythrocytic Hypoplasia: A Recovery During Cobalt Therapy (Report of Two Cases and Review of Literature), Acta haemat. **9**: 153, 1953.
27. Smith, C. H.: The Anemias of Early Infancy, J. PEDIAT. **16**: 375, 1940.
28. Smith, C. H.: Chronic Congenital Aregenerative Anemia (Pure Red-cell Anemia) Associated With Iso-immunization by the Blood Group Factor "A," Blood **4**: 697, 1949.
29. Smith, C. H.: Hypoplastic and Aplastic Anemias of Infancy and Childhood: With a Consideration of the Syndrome of Nonhemolytic Anemia of the Newborn, J. PEDIAT. **43**: 457, 1953.
30. Smith, C. H., Schulman, I., Ando, R. E., and Stern, G.: Studies in Mediterranean (Cooley's) Anemia. II. The Suppression of Hematopoiesis by Transfusion, Blood **10**: 707, 1955.
31. Tsai, S. Y., and Levin, W. C.: Chronic Erythrocytic Hypoplasia in Adults. Review of Literature and Report of a Case, Am. J. Med. **22**: 322, 1957.
32. Vogel, P., and Bassen, F. A.: Sternal Marrow of Children in Normal and in Pathological States, Am. J. Dis. Child. **57**: 245, 1939.
33. Vogel, P., Erf, L. A., and Rosenthal, N.: Hematological Observations on Bone Marrow Obtained by Sternal Puncture, Am. J. Clin. Path. **7**: 436, 1937.

MEGALOBLASTIC ANEMIA IN INFANCY

III. Clinical Considerations and Analysis

A. Leonard Luhby, M.D.
New York, N. Y.

MEGALOBLASTIC anemia during infancy is more than an anemia. It is a syndrome that is often characterized by failure to gain weight, loss of appetite, reduced ability to cope with infection, and hemorrhagic phenomena. Although the full-blown clinical pattern has become less common during the past several years, the clinical importance of "megaloblastic anemia" is not to be deprecated. Patients with this syndrome are potentially more seriously ill than the degree of anemia would lead one to believe.

It is the purpose of this article to present briefly clinical and experimental studies of interest to the general medical and pediatric practitioner. The majority of the data is from patients studied at the Children's Hospital, Columbus, Ohio, 1947 to 1950. Some of the findings were previously reported.[1] It is an interesting but still unexplained sidelight that this disorder is more common in the Midwest and certain areas of the Far West than in the eastern part of the United States.

CLINICAL CHARACTERISTICS

The syndrome of megaloblastic anemia in infancy is an acute disorder of temporary nature, rarely chronic, if properly treated, and is not to be confused with so-called juvenile pernicious anemia, which may commence in infancy, but is a chronic, usually lifelong, disease.

The age incidence of the megaloblastic anemia syndrome is shown in Fig. 1. The youngest patients were 6 to 8 weeks of age, the oldest in the series reviewed, 17 months. Although the mean age may be 7 to 8 months, it is important to observe that a considerable number of cases occur in very early infancy, 2 to 4 months of age, when other nutritional anemias are uncommon.

The onset of the disorder is insidious. Pallor is the most universal clinical feature. When the syndrome is established, there is lack of appetite, failure of continued weight gain, crankiness, weakness, and inability to sit up unaided, when the latter had been possible previously. Petechial skin and mucous membrane hemorrhages occur in a small percentage of cases and occasionally lead to an initial diagnosis of thrombocytopenic purpura. If scurvy is also present, there may be refusal to use the legs and bleeding phenomena. There were 3 cases of scurvy in our series of 27 patients between 1947 and 1952. Scurvy, however, is much less frequently associated with the megaloblastic anemia syndrome in the United

From the Department of Pediatrics, New York Medical College.

States today than previously. Fever and evidence of infection is common, either preceding the development of pallor or apparently simultaneous with or following it.

Since the reduction in the incidence of this syndrome in this country due to the introduction of ascorbic acid fortification of modified powdered and liquid milks for infant feeding, the clinical history that should alert the physician's suspicions of megaloblastic anemia today is the development of pallor, persistence of moderate anorexia, and failure to gain weight in an infant who, recovering from a severe or protracted infection or diarrhea, would ordinarily be expected to begin to thrive.

The most common findings on physical examination are pallor, the extent varying with the degree of anemia, and hepatomegaly. A yellow cast may be present in the very anemic patient, but there is no true jaundice unless it is caused by another coexisting disease process. Splenomegaly is not infrequent, but it is unusual for the spleen to be greatly enlarged. Petechiae and ecchymoses are usually present in the more severely affected patients or in those who have been unrecognized as having megaloblastic anemia and have had blood transfusions. In latter instances, the underlying metabolic deficiency is not corrected. With the further progression of the disease, thrombocytopenia and purpuric phenomena develop. If scurvy is present, the lower extremities may be flexed and tender, due to subperiosteal bleeding. Central nervous system disease, often a feature of pernicious anemia,

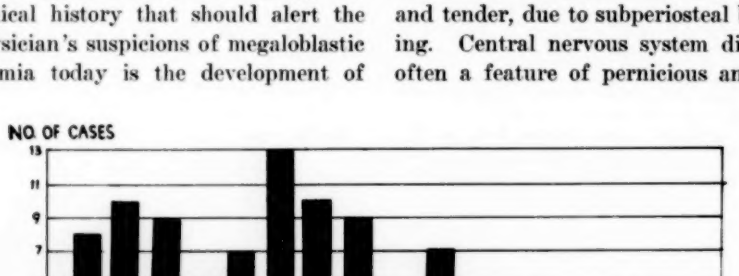


Fig. 1.—Relation of onset of megaloblastic anemia syndrome to age of infants; compiled from cases reported in United States between 1946 and 1952, including the author's 27.

was not present in any of our patients and has not been reported anywhere except in Italy.[2, 3] The fact that many of our patients would not sit up unsupported, although previously able to, was attributed to the generalized weakness accompanying the syndrome rather than to neurologic involvement.

Although there is probably no constitutional predilection toward the development of the megaloblastic anemia syndrome, we and others have observed two general types. One is the blond, blue-eyed, very obese infant, usually beyond 6 to 7 months of age. This type of baby develops the syndrome primarily on a dietary basis.

He is well fed "calorically" but has a poorly balanced diet, chiefly consisting of carbohydrates. The other type, usually but not entirely limited to infants less than 6 months old, is the thin, scrawny, but long infant who has probably just recovered from a long siege of infection or diarrhea. This infant develops the syndrome usually because he cannot cope with the increased metabolic demands of infection which at the same time limits his intake of essential nutrients.

LABORATORY FEATURES

The anemia may be mild or severe. When severe, the hemoglobin can be among the lowest encountered in clinical medicine, 1.5 Gm. per cent, and the red blood cell count less than one million per cubic millimeter. The red blood cell morphology may show considerable variation in shape and size. The over-all pattern is usually one of normocytosis and normal or slight hypochromia. Absolute macrocytosis and hyperchromia, as judged from the mean corpuscular volume and the mean corpuscular hemoglobin measurements, are usually "masked" by the microcytosis and slight hypochromia present in infancy. However, occasional or moderate numbers of macrocytes may be seen on smear. Nucleated red blood cells may be present in the peripheral blood of moderately to severely affected infants. These occasionally show megaloblastic nuclear features.

The white cell blood count is often at leukopenic levels in the severe case uncomplicated by infection. Because infection is such a frequent accompaniment of the syndrome, however, leukopenia is clinically uncommon. Total white cell blood counts usually vary from 7,000 to 20,000 per cubic millimeter. Increased or abnormal segmentation and moderate increase in size of the neutrophils is characteristic of the disorder. These features may be present in the infant only moderately affected, whether there is a leukopenia or leukocytosis, and often may give rise to the first suspicions of the syndrome to the alert laboratory technician.

Thrombocytopenia, an inherent part of the disorder, is often present in a mild degree in the moderately involved infant, for the cellular "megaloblastic" defect in the syndrome affects all the blood and body cells. In severe cases, platelet counts less than 80,000 per cubic millimeter are common.

Bone marrow aspiration smear, examined after staining with Wright's or Jenner-Giemsa technique, is the most reliable diagnostic test. The marrow is usually cellular and has an erythroid hyperplasia with an increase in the more immature erythroid cellular forms. The megaloblastic erythroid abnormalities are present throughout the developing red cell precursor series but are more easily recognized at the "erythroblast" rather than the earlier "proerythroblast" or later "normoblast" stage. Briefly, the nuclear chromatin occurs in larger and coarser masses; there are abnormal nuclear lobulation, budding, fragmentation, and atypical mitosis. Also there are characteristic morphologic abnormalities in the myeloid series beyond the myeloblast stage. The more common abnormalities in the myelocytes and metamyelocytes are early indentation and lobulation of the

nucleus, retention of cytoplasmic immaturity, vacuolization, and increase in size of the cells. In the mature polymorphonuclear leukocytes, the nuclear abnormality results in increased and atypical nuclear segmentation. The megakaryocytes in severe cases also show an increase in size and abnormal and increased nuclear lobulation.

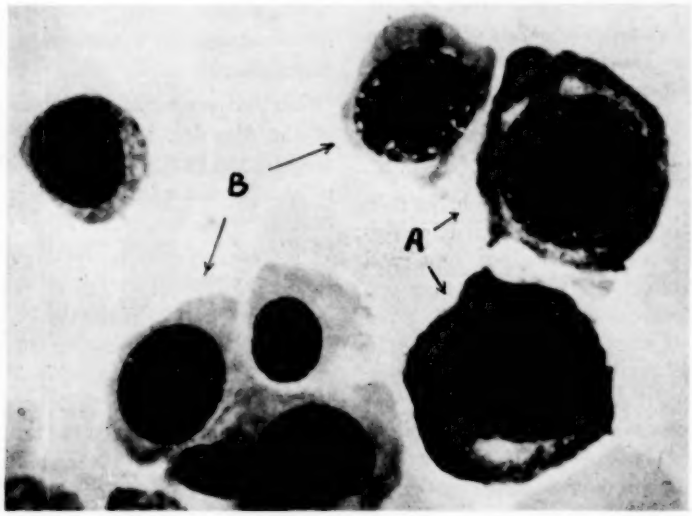


Fig. 2.—"Intermediate" megaloblasts in the erythroid series in a patient with moderate megaloblastic anemia. Note the more typical megaloblastic chromatin structure in the early erythroblasts (*A*) and the less well-defined changes in the normoblasts (*B*). Cell at upper left is small lymphocyte.

The foregoing features represent the so-called "megaloblastic bone marrow pattern." It is pathognomonic of the blood disorder associated with an absence of either folic acid, vitamin B_{12}, or other related factors. Among the many excellent illustrations in the literature of the blood and marrow cellular abnormalities in the megaloblastic anemia syndrome are those in Zuelzer and Ogden's classic report.[4]

Today, severe classic megaloblastic marrow patterns are uncommon in this country. Hematologists, however, recognize an intermediate pattern in which macrometamyelocytes with abnormal nuclear lobulation and megaloblastic erythroid precursors (Fig. 2) with less pronounced nuclear chromatin changes are present. This pattern represents a state of lesser "deficiency" that is currently more common, particularly following diarrhea or infection. Interestingly, the myeloid cells are usually the first to show megaloblastic changes in the early or incipient stages of the syndrome. The incidence of megaloblastic anemia of infancy in different clinics often varies in direct proportion to the frequency with which the aspirated bone marrow of all even mildly anemic infants is examined and the recognition of the "intermediate megaloblastic pattern" as being diagnostic of early megaloblastic anemia.

Evaluation of gastric function includes acidity measurements which usually reveal a normal fasting and histamine-reactive pattern. Occasional patients show a fasting and histamine-refractory hypo- or anacidity. We found gastric "intrinsic factor" output to be normal in several patients with megaloblastic anemia of infancy.[5] More recently, it has been shown also that the mucoprotein content of the gastric juice, which roughly parallels the "intrinsic factor" activity, is low during the neonatal period but rapidly approaches adult concentrations in normal infants during the first few months of life.[6]

Stools may be light yellow, greasy, and somewhat bulky if the syndrome has been present for some time. We noticed steatorrhea develop in infants in whom the disease was only partially corrected for several weeks when the therapeutic effectiveness of various agents was being evaluated. Fat absorption studies were not done in these infants, but impaired gastrointestinal fat absorption is part of the abnormal physiology associated with folic acid deficiency, e.g., tropical sprue. The steatorrhea in our patient improved completely following folic acid therapy. Similar findings have been reported abroad.[7]

Roentgenograms of the long bones are usually normal. A moderate number of cases show osteoporosis and a few, the early "ground-glass" appearance suggestive of subclinical scurvy.

Newer Diagnostic Aids.—In recent years several new tests have become available to aid the evaluation of folic acid and vitamin B_{12} nutrition.

Serum folic acid levels have not been of clinical usefulness to date because assay techniques with sufficient specificity have not been available. This deficiency has led to our interest in formiminoglutamic acid excretion as an indication of folic acid nutriture.

Formiminoglutamic acid urinary excretion parallels the development of folic acid deficiency in the rat under experimental conditions.[8] This compound, a normal intermediate in amino acid metabolism, requires the metabolically active form of folic acid, tetrahydrofolic acid,[9] for its further degradation to glutamic acid. When insufficient tetrahydrofolic acid is available, formiminoglutamic acid (FIGLU) accumulates. Luhby and Broquist[10, 11] have shown that formiminoglutamic acid is also excreted in folic acid deficiency states in man. Subsequent studies[12] revealed that urinary FIGLU is a useful indicator of clinical folic acid deficiency. Formiminoglutamic acid may be assayed by enzymatic, bacteriologic, or chromatographic techniques. Normal values range from 0 to 30 μg per cubic centimeter of urine.[13] We have found increased urinary FIGLU in megaloblastic anemia of infancy.

Serum vitamin B_{12} levels reflect for practical purposes the state of vitamin B_{12} nutrition, when liver disease or chronic leukemia can be ruled out. Measurement of the very minute amounts of vitamin B_{12} present in serum requires microbiological assay with such organisms as *Ochromonas malhemensis, Euglena gracilis,* or *Lactobacillus leichmannii.* Each microorganism has special characteristics which make it useful for different purposes. Ochromonas has the greatest specificity. It will not respond to "pseudovitamin B_{12}" present in the serum. Euglena is the most

sensitive. It can detect 0.5 μμg of vitamin B_{12} per cubic centimeter. Microbiological assay with either *O. malhemensis* or *E. gracilis* as the test organism, however, has the disadvantage of requiring 5 to 7 days for completion. The *L. leichmannii* assay has the advantage of speed, requiring an incubation period of only 24 hours, but lacks the specificity and sensitivity of the other two.

The vitamin B_{12} serum level in the "normal" young adult varies somewhat with the assay technique but on the average is in the range of 250 to 550 μμg per cubic centimeter. Extensive studies of vitamin B_{12} levels in the immediate neonatal period show values ranging from 200 to 1,300 μμg per cubic centimeter, with a mean of 560.[14] Vitamin B_{12} levels in later infancy and early childhood have not been extensively studied. One report indicates a range of 300 to 900 μμg.[15] I am unaware of studies of the serum vitamin B_{12} in infants with megaloblastic anemia syndrome. In folic acid deficiency states, such as nutritional macrocytic anemia, serum vitamin B_{12} levels are usually within normal range. In pernicious anemia, the serum vitamin B_{12} levels are low, frequently 100 μμg per cubic centimeter or less.

Gastrointestinal absorption of vitamin B_{12} is evaluated by employing radioactive vitamin B_{12} in either the urinary excretion test proposed by Schilling[16] or the hepatic uptake test proposed by Glass and his associates.[17] The urinary excretion test is carried out by administering a small oral dose of radioactive vitamin B_{12}, usually 2 μg, containing 0.5 μc of radioactivity, followed by an injection of 1,000 μg of nonradioactive vitamin B_{12}, either immediately or within 2 hours of the oral dose. The patient's urine is collected for the next 24- or 48-hour period and the radioactivity in each 24-hour aliquot determined. Approximately 10 per cent of the radioactivity administered is recovered in the urine within the first 24 hours and an additional 5 per cent during the second 24 hours in normal individuals. In pernicious anemia, the average recovery is 0.7 per cent in the first 24 hours and 0.2 per cent in the second 24 hours. Following the administration of intrinsic factor to patients with pernicious anemia, the excretion may increase to approximately 6 per cent in the first 24 hours and 2.5 per cent in the second 24 hours.[18]

The hepatic uptake test employs the oral administration of 1.0 to 2.0 μg radioactive vitamin B_{12} orally, containing 1.0 to 0.5 μc of radioactivity, first without and then with intrinsic factor.[19] Five to seven days following the test, the radioactivity over the patient's liver area is scanned externally with a probe-type scintillation detector. In the individual with normal gastrointestinal absorption, the hepatic radioactivity at this time ranges from 150 to 350 counts per microcurie administered. In pernicious anemia or with the malabsorption syndromes, the radioactivity detected over the liver is considerably less. Administration of radioactive B_{12} with intrinsic factor in pernicious anemia increases the radioactivity over the hepatic area toward normal. Neither of these tests had been developed when severe megaloblastic anemia syndrome in infants was more prevalent in this country.

DIFFERENTIAL DIAGNOSIS

Differential diagnosis of the megaloblastic anemia syndrome includes the more common iron-deficiency anemia of infancy, the syndrome of "iron deficiency anemia, hypocupremia and hypoproteinemia" (although edema is rare in megaloblastic anemia, 2 of our 27 patients developed a mild pitting edema), thrombocytopenic purpura, aplastic anemia, and juvenile pernicious anemia. In the latter, the serum vitamin B_{12} level and evaluation of intrinsic factor activity and gastrointestinal absorption by either the urinary excretion test or the hepatic uptake test, described above, are the most clinically useful means of differentiating it from the megaloblastic anemia syndrome. Occasionally, patients with celiac disease develop megaloblastic anemia, although it is less frequent than expected.

INCIDENCE ELSEWHERE IN THE WORLD

Although the megaloblastic anemia syndrome has become less common in infants in this country, it is currently an important problem elsewhere in the world. The increased incidence in other countries may be due partially to an increase in the use of bone marrow aspirations in the differential diagnosis of anemias in infancy. Interesting reports of this syndrome have appeared recently from Durban, South Africa, by Adams[20] and Walt, Holman, and Hendrickse.[21] Walt and his colleagues describe 42 cases of megaloblastic anemia which represented 5.4 per cent of all pediatric admissions to their hospital in a one-year period. Lien-Keng, Poesponegoro, and Hin[22, 22a] found approximately 25 per cent of their patients with kwashiorkor and anemia to have the megaloblastic syndrome. In an extensive study of infants with anemia at the University College of the West Indies Hospital, Jamaica, MacIver and Black[23] found 20 cases of the megaloblastic syndrome in 1957. This represented 6.3 per cent of all admissions to their pediatric ward for the period.

TABLE I. DIETARY HISTORY PRECEDING MEGALOBLASTIC ANEMIA SYNDROME IN INFANCY

AUTHOR'S CASES: 1947-1952					
SOLID FOOD			ASCORBIC ACID		
	NUMBER OF CASES	PER CENT		NUMBER OF CASES	PER CENT
Well taken	3	11	Good intake	3	8
Poorly taken	4*	15	Occasional intake	4*	16
Not offered, refused, or vomited	20	74	Not offered	18	76
Totals	27			25	

*Not same patients.

ETIOLOGY

The pathologic physiology in this syndrome is basically a derangement of nucleoprotein metabolism. Because there are many steps involved, theoretically a break in any one may give rise to a "megaloblastic anemia." The etiology and pathogenesis of this syndrome may thus differ in different individuals and in different areas of the world.

In this country the first large series of infants with this anemia was reported from Detroit by Zuelzer and Ogden.[4] Soon thereafter a smaller but well-studied series was reported from Minneapolis by Aldrich and Nelson.[24] These reports stimulated our clinical and experimental studies in human infants with the disease in Columbus, Ohio. May, Nelson, Lowe, and their co-workers[29] followed the lead of Aldrich and Nelson's study. They were particularly impressed with the very low vitamin C intake of their patients and set out to reproduce the syndrome in monkeys.

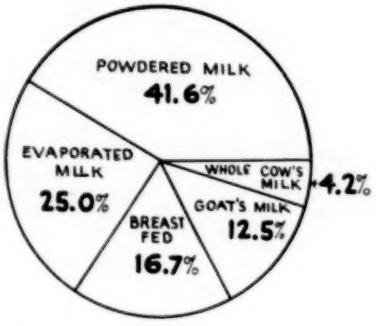


Fig. 3.—Types of milk formulas fed prior to onset of megaloblastic anemia syndrome in 27 patients, 1947 to 1952.

Role of Diet in Clinical Etiology.—An analysis of the previous dietary history of our patients (Table I) clearly shows that in the great majority the intake was extremely poor for not only ascorbic acid but solid foods as well.

Analysis of the types of milk used for the formulas of our patients (Fig. 3) indicates the relatively high percentage, 41.6, of modified powdered milk, which was similar to the experience of others at that time. As for goat's milk, it has long been implicated in the American and German literature in the etiology of "macrocytic anemia" in infants which resembles pernicious anemia and responds to liver extract.[25, 26, 27] Bachman,[28] however, was the first in this country to recognize the megoloblastic anemia syndrome as a separate clinical entity in an infant with a poor diet and episodes of vomiting and diarrhea and to differentiate it from Addisonian pernicious anemia.

A comparison of the type of milk fed to the age of occurrence of the syndrome (Fig. 4) reveals interesting relationships that are not widely known. Note that the peak of incidence for infants fed powdered or goat's milk formulas was approximately 5 to 9 months of age, although cases occurred earlier and later. As will be seen below, this is, in general, the same group of infants who probably had a mild or incipient anemia which was "precipitated" or made worse by infection. Stated another way, this is the group of infants in whom moderate or repeated mild infections could not be tolerated without development of hematologic and metabolic abnormality. It is also of interest that most of the infants who were breast fed developed the anemia after 10 months of age. This would imply that although breast milk alone may not be totally adequate to prevent megaloblastic anemia, it contained more of the necessary nutrients than powdered or goat's milk because it permitted the infants to thrive for a longer time without precipitation of the syndrome. The age incidence of infants fed an evaporated milk formula corresponded, in general, to the

age of onset if severe infection, group 1 *a* in Fig. 5, was plotted as the chief precipitating factor.

Despite considerable research to date, the deficiencies in these various milks which contribute to precipitation of the megaloblastic anemia syndrome is not completely understood.

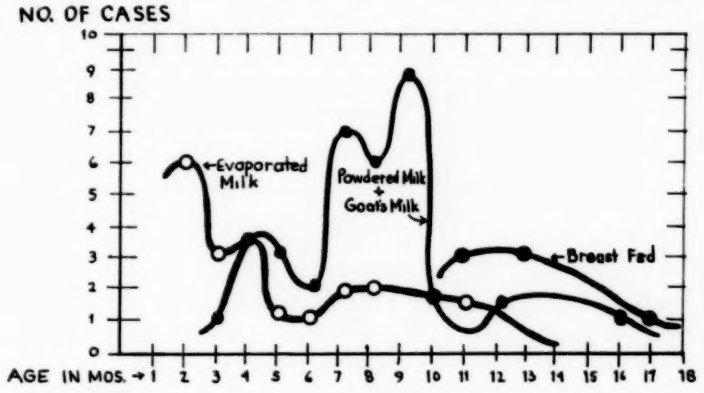
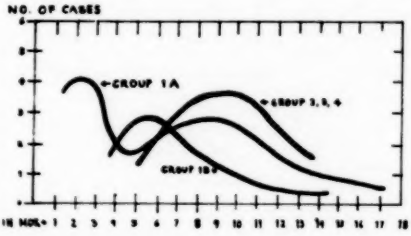


Fig. 4.—Relation of type of milk formula fed to the age of occurrence of megaloblastic anemia syndrome in the 92 cases analyzed for Fig. 1.



GROUP	PRINCIPAL CAUSE OF ANEMIA	PEAK of INCIDENCE
1 *a*	Severe infection.	2–4 mos.
1 *b*	Poor diet + precipitating infection.	4–7 "
2, 3 and 4	Inadequate diet without significant infection.	8–12 "

Fig. 5.—Relation of age at onset of megaloblastic anemia syndrome to principal cause in author's 27 cases.

One factor is apparent. Milks from various species are extremely poor sources of folic acid (Table II). The levels in any type of milk are considerably below the probable minimal daily requirements. Although the minimal daily requirement of folic acid for man has not yet been established, some experimental evidence in man and monkeys indicates it may be in the range of 100 to 200 μg daily.

The natural ascorbic acid content of milk is only fair and varies considerably with the vitamin C intake of the lactating female. Cow's milk is an excellent source of vitamin B_{12}; goat and human milk are low. In addition, folic acid, like ascorbic acid, is labile

and destroyed by heat and light. Thus, cooking and commercial preparative processes are prone to lower further the already subminimal content of folic acid and the minimal levels of ascorbic acid naturally present in milk.

TABLE II. FOLIC ACID, VITAMIN B_{12} AND ASCORBIC ACID CONTENT OF MILK*

	FOLIC ACID (μg/L.)	VITAMIN B_{12} (μg/L.)	VITAMIN C (mg./L.)
Cow milk	2.0 (1-5)	5.6 (0.7-11.5)	16 (2-31)
Human milk			
(a) Colostrum (1 to 5 days)	0.5 (0.1-1.5)	0.4 (0.1-1.5)	44 (4-104)
(b) Mature	2.0 (1-3.6)	0.2	43 (0-112)
Goat milk	0.3 (0-1.6)	0.2 (0-1.4)	14 (0-32)

*Handbook of Biological Data, William S. Spector, Editor. Philadelphia, 1956, W. B. Saunders Co., p. 50.

our 27 cases (Table III) showed that in approximately one quarter of these (group 1a) severe infection appears to be the major clinical etiological factor. In another small number (group 1b), infection played a contributing role in limiting the dietary intake. Thus, in about one third of the patients, infection was important in the etiological factors leading to development of the syndrome. In group 3, it is difficult to evaluate the primary role of infection. Here it was obvious the patients were mildly anemic before infection occurred and that the infectious process merely worsened the pre-existing condition. In group 4, infection definitely followed the anemia. Only 18.5 per cent of the patients, group 2, had no com-

TABLE III. RELATIVE RELATION OF INFECTION AND DIET IN CLINICAL ETIOLOGY OF MEGALOBLASTIC ANEMIA SYNDROME IN INFANCY

AUTHOR'S CASES: 1947-1952			
	PRINCIPAL CLINICAL CAUSE	NUMBER OF CASES	PER CENT
Group 1a	Severe and acute infection; good or fair preceding dietary intake	7	26.0
Group 1b	Moderate but chronic infection; with obvious infection-induced poor dietary intake	2	7.5
Group 2	Poor diet taken due to mother's ignorance, infant's refusal of feedings, persistent vomiting, allergy, or other cause; no infection	5	18.5
Group 3	Poor preceding diet with "precipitating" or simultaneous infection(s)	9	33.3
Group 4	Poor preceding diet with infection definitely following onset of anemia	4	14.7
Totals		27	100.0

In contrast, vitamin B_{12} is not affected by such procedures.

Role of Infection in Clinical Etiology.—It was apparent from a review of our cases, as well as those of others, that dietary inadequacies were not the sole factors in the etiology of the megaloblastic anemia syndrome. A small but significant number of patients had a very good preceding diet and others, a "fair" diet.

An analysis of the relation of infection to the development of anemia in plicating infections either before or after development of the anemia. The clinical types of infection in our patients is shown in Table IV.

An analysis of the relation between the age of occurrence and whether infection or dietary inadequacy was the principal cause of the syndrome is shown in Fig. 5. Infection is invariably the most important precipitating factor in the younger, 2- to 4-month, age group. In the older age groups, particularly after 8 months, the

"physiological stress" of infection must have been better tolerated when the preceding diet was adequate. Conversely, this clinical evidence indicates the poor reserves the younger infant possesses to prevent the precipitation of the megaloblastic syndrome by infection. The peak incidence of this syndrome in this country has now shifted toward the earlier months of life, reflecting reduction in groups 2, 3, and 4 (Table III) through improved dietary regimens recommended during the past 7 to 8 years.

TABLE IV. CLINICAL TYPES OF INFECTION ASSOCIATED WITH MEGALOBLASTIC ANEMIA SYNDROME IN INFANCY

27 CASES: 1947-1952		
	NUMBER OF INSTANCES	PER CENT
Diarrhea, severe, usually prolonged	6	27.6
Pneumonia, moderate, frequently chronic	5	23.0
Upper respiratory infection, often multiple	4	18.3
Pyelitis, chronic	2	9.1
Skin abscess, large, chronic	1	4.5
Meningitis, acute, pneumococcal	1	4.5
Septicemia, severe, acute	1	4.5
Stomatitis, chronic	1	4.5
Conjunctivitis, purulent, subacute	1	4.5

The relative roles of diet and infection in the production of the megaloblastic anemia syndrome in the United States today is, in our experience, represented by that found in patients of groups 1a and 1b, Table III.

EXPERIMENTAL STUDIES

It is not within the scope of this report to extensively review the many excellent experimental studies which led to our present-day understanding of the etiology and pathogenesis of the megaloblastic anemia syndrome.

The relative roles of folic acid and vitamin B_{12} in the pathogenesis of megaloblastic anemia in human infants were partially elucidated from experiments such as the four shown in Fig. 6. Luhby and Wheeler[1] were the first to demonstrate that infants with this disorder did not respond to vitamin B_{12}. Because of the excellent response to small doses of folic acid, it became apparent that folic acid was the chief deficient nutrient. When responses were obtained with vitamin B_{12}, this was due to the "potentiating" or "sparing" action of large doses of vitamin B_{12} in metabolic processes where folic acid was in short supply. An experiment such as Fig. 6*A* indicated that a possible temporary deficiency of gastric "intrinsic factor" was not the cause of the disorder because the patient did not respond to the administration of beef plus normal gastric juice, a regimen which produced a response in a patient with pernicious anemia.

Other experiments, to be reported in detail elsewhere, demonstrated that large doses of ascorbic acid, totaling 3,000 mg. given with a small 0.3 mg. dose of folic acid did not correct the megaloblastic anemia in an infant who was on this regimen for over 2 months. Thus, once the anemia is developed, the "folic acid deficiency" is so severe that the potentiating effect of ascorbic acid is negligible.

May, Nelson, Lowe, and their coworkers[29] in an excellent series of experiments in monkeys showed that these animals would develop scurvy and then megaloblastic anemia after

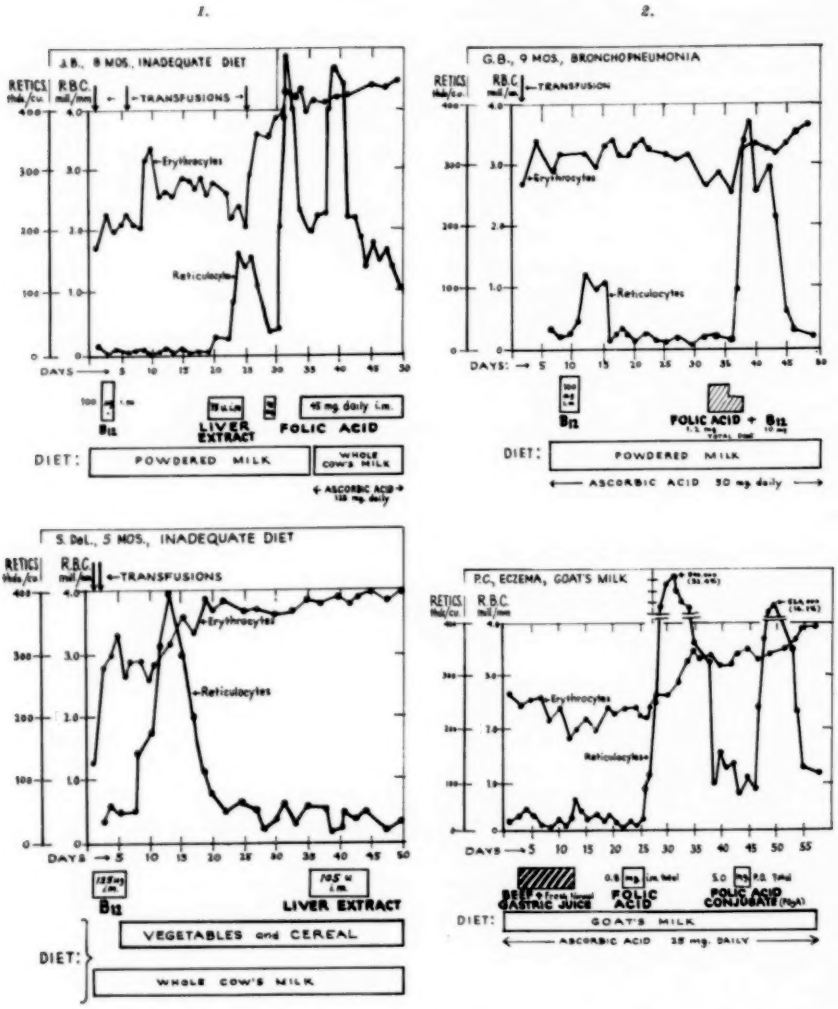


Fig. 6.—Response of megaloblastic anemia to various therapeutic regimens in the same patient. Diet during experimental period is indicated.

1, Therapeutic failure of 100 μg vitamin B_{12} intramuscularly; submaximal response to 75 units of refined liver extract intramuscularly; excellent and complete response to subsequent folic acid orally.

2, Partial but incomplete therapeutic response to a single dose of 100 μg of vitamin B_{12} intramuscularly, followed by a daily oral dose of 50 mg. ascorbic acid; excellent response to subsequent minimal 1.2 mg. dose of folic acid intramuscularly, given simultaneously with 10 μg vitamin B_{12} parenterally.

3, Failure of response to 125 μg of vitamin B_{12} parenterally; excellent response following addition of vegetables and cereal on fifth day after vitamin B_{12}; no further effect of liver extract given 1 month later.

4, Therapeutic failure to classical Castle experiment: ground beef (containing "extrinsic factor" now known to be identical with vitamin B_{12}) plus normal gastric juice (containing intrinsic factor" necessary for gastrointestinal absorption of vitamin B_{12}); excellent and complete response to subsequent administration of folic acid and then folic acid conjugate (akin to folic acid as it occurs in foodstuffs).

(Adapted from Luhby and Wheeler.[1])

periods of from 4 to 6 months if fed a diet containing primarily a commercial modified powdered milk without additional ascorbic acid. Addition of ascorbic acid to the diet at the start of the experiment prevented the appearance of scurvy and megaloblastic anemia as long as the animals remained free of infection. Later, they

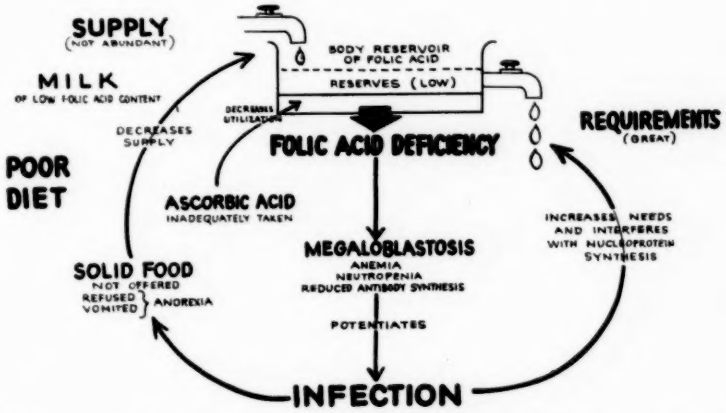


Fig. 7.—Diagrammatic representation of the relationships of infection and diet in the causation of the megaloblastic anemia syndrome in infants.

demonstrated[30] that infection or turpentine abscesses could precipitate megaloblastic anemia in monkeys fed the same milk diet even *with* the addition of 50 mg. of ascorbic acid daily.

Such animal experiments demonstrated the importance of ascorbic acid in potentiating minimal amounts of folic acid and the ability of infection to deplete the body stores of folic acid despite adequate vitamin C.

THEORY OF PATHOGENESIS

A deficiency of folic acid, vitamin B_{12}, and other nutrients can lead to the development of the megaloblastic anemia syndrome. It appears clear from clinical and animal experiments that the basic defect in the overwhelming majority of instances of megaloblastic anemia in infancy and childhood is due to a "deficiency of folic acid" or its equivalent. Those due to a deficiency of vitamin B_{12} are rare. The folic acid deficiency may arise from gastrointestinal malabsorption as occurs in the few instances of megaloblastic anemia associated with celiac disease, but in most cases it is a "nutritional folic acid deficiency" brought on by either a poor supply or increased requirements. The role of protein in the diet in the development of megaloblastic anemia is under study.

The relationships between supply and demand in the production of megaloblastic anemia are illustrated diagrammatically in Fig. 7. The average infant's reserve of folic acid is probably at the lower limit of normal. If it decreases below a certain level, megaloblastic anemia develops because there is insufficient folic acid for conversion to tetrahydrofolic acid which is necessary as a coenzyme for amino acid metabolism leading to nucleoprotein and new cellular formation.

"Citrovorum factor," or folinic acid, is not the metabolically active form of folic acid[9] as was previously believed. The infant receives most of his folic acid supply from solid foods, for milks are poor sources. When plentiful, ascorbic acid aids in reduction of folic acid to tetrahydrofolic and may spare it in certain amino acid (tyrosine) metabolic processes. Ascorbic acid will not, however, replace the requirement for folic acid. Infection increases the requirements for all the water-soluble vitamins, including folic acid and vitamin C. Infection will result in the lowering of their level if the supply is unchanged. In addition, folic acid deficiency leads to neutropenia and reduced antibody synthesis[31] which may potentiate or give rise to further infection.

MANAGEMENT

Treatment of megaloblastic anemia of infancy must be considered from the point of view of specific and supplementary therapy.

There is only one specific therapy, *folic acid*. Ten to thirty milligrams daily, orally if the infant will retain it, intramuscularly if he will not, until the blood count returns to normal is recommended. This is all that is necessary for a prompt, complete, and lasting cure of all aspects of the syndrome. Liver extract, refined, 10 to 15 units intramuscularly daily produces fair to good responses, but these responses are not as good as with folic acid. Crystalline vitamin B_{12}, 50 to 100 μg intramuscularly daily, produces responses in some patients but is not reliable (Fig. 6). Because of the possibility of partial response or complete failure, we consider vitamin B_{12} *contraindicated* for the clinical treatment of this condition.

Supplementary therapy includes transfusion, dietary, and anti-infection measures. If the patient's hemoglobin level is 5.0 Gm. per cent or less, transfusion to raise it is indicated. As in most clinical nutritional anemias, it is rare for only one nutrient to be in short supply. Therefore, supplements with ascorbic acid, particularly if there is frank or roentgen evidence of scurvy, and the other B vitamins may be helpful and is good medical practice. It should be emphasized, however, that unless there is evidence of another vitamin deficiency in addition to megaloblastic anemia, these supplements are not essential.

Measures to counteract infection should be instituted as the situation requires. The presence of neutropenia does not contraindicate the use of antibiotics which occasionally result in a blood dyscrasia if the particular antibiotic is the one of choice for the infection present.

PROPHYLAXIS

To prevent megaloblastic anemia it is important the infant receive adequate supplies of folic acid beginning early in life. These are ordinarily obtained from vegetables, egg, and meat. Good vitamin C intake, 30 to 50 mg. daily, is also advisable.

When the infant's food intake is reduced for a considerable period of time or if there is severe or prolonged infection, the physician should be alert to the infant's nutritional requirements. Judicious and appropriate administration of B vitamins, including folic acid and ascorbic acid, during such periods are indicated.

SUMMARY

Megaloblastic anemia syndrome in infancy is an important nutritional deficiency disorder usually precipitated in young infants by a combination of infection and dietary inadequacies. Severe or prolonged infection or diarrhea with coexisting poor food intake is the most important clinical etiological factor today. The disorder responds promptly, completely, and permanently to folic acid administration.

These studies were made possible by the encouragement and advice of Dr. W. E. Wheeler, Dr. E. H. Baxter, and Dr. C. A. Doan of the Ohio State University College of Medicine. The untiring technical assistance of Mrs. Dorothy Conrad Baxter and Miss D. Crane, as well as the resident and nursing staffs of the Children's Hospital, Columbus, Ohio, were invaluable. The studies in Columbus were supported in part by the Ross Laboratories through the courtesy of Mr. J. J. Quilligan and Dave Cox and the Blood Grouping Laboratory of Boston, through the courtesy of Dr. L. K. Diamond. The Playtex Park Research Institute and the National Vitamin Foundation supported in part the studies in New York.

REFERENCES

1. Luhby, A. L., and Wheeler, W. E.: Megaloblastic Anemia of Infancy. II. Failure of Response to Vitamin B_{12} and Metabolic Role of Folic Acid and Vitamin C, Ohio State Univ. Health Center J. **3**: 1, 1949.
2. Pecorella, F., Burgio, G. R., and Aversa, T.: Observazioni cliniche ed etiopatogenetiche su 18 casi di anemia perniciosiforme megaloblastica nel latante, Siv. Clin. pediat. **45**: 65, 1947.
3. Amato, M.: Die neurologischen Erscheinungen der reversiblen Megaloblastenanämie im Kleinkindesalter, Helvet. paediat. acta **7**: 291, 1952.
4. Zuelzer, W. W., and Ogden, F. N.: Megaloblastic Anemia of Infancy, Am. J. Dis. Child. **71**: 211, 1946.
5. Luhby, A. L.: Megaloblastic Anemia of Infancy. I. Extrinsic-Intrinsic Factor Relationships, Ohio State Univ. Health Center J. **2**: 35, 1948.
6. Luhby, A. L., Glass, G. B. J., and Slobody, L. B.: Gastric Mucoprotein Secretion in Infants and Children, A. M. A. Am. J. Dis. Child. **88**: 517, 1954.
7. Orsini, M., and Maggioni, G.: Megaloblastic Anemia and Steatorrhea in Breast Fed Infants Cured With Folic Acid, Hematologica **36**: 877, 1952.
8. Bakerman, H., Silverman, M., and Daft, F.: Influence of Succinylsulfathiazole and Folic Acid on Glutamic Acid Excretion, J. Biol. Chem. **188**: 117, 1951.
9. Greenberg, G. R., and Jaenicke, L.: On the Activation of the 1-Carbon Unit for the Biosynthesis of Purine Nucleotides, Ciba Foundation Symposium, The Chemistry and Biology of Purines, Boston, 1957, Little, Brown & Co., p. 204.
10. Luhby, A. L., and Broquist, H. P.: Urinary Formiminoglutamic Acid, a Biochemical Test for Folic Acid Deficiency, A. M. A. J. Dis. Child. **94**: 553, 1957.
11. Broquist, H., and Luhby, A. L.: Detection and Isolation of Formiminoglutamic Acid From Urine in Folic Acid Deficiency in Humans, Proc. Soc. Exper. Biol. & Med. **100**: 349, 1959.
12. Luhby, A. L., Cooperman, J. M., Teller, D. N., and Donnenfeld, A. M.: Excretion of Formiminoglutamic Acid in Folic Acid Deficiency States, J. Clin. Invest. **37**: 915, 1958.
13. Luhby, A. L., Cooperman, J. M., and Teller, D. N.: Unpublished Data.
14. Boger, W. P., Bayne, G. M., Wright, L. D., and Beck, G. D.: Differential Serum Vitamin B_{12} Concentrations in Mothers and Infants, New England J. Med. **256**: 1085, 1957.
15. Killander, A. B.: Serum Vitamin B_{12} Levels at Various Ages, Acta paediat. **46**: 585, 1957.
16. Schilling, R. F.: Intrinsic Factor Studies. II. Effect of Gastric Juice on Urinary Excretion of Radioactivity After Oral Administration of Radioactive Vitamin B_{12}, J. Lab. & Clin. Med. **42**: 860, 1953.
17. Glass, G. B. J., Boyd, L. J., Gellin, G. A., and Stephanson, L.: Uptake of Radioactive Vitamin B_{12} by the Liver in Humans: Test for Measurement of Intestinal Adsorption of Vitamin B_{12} and Intrinsic Factor Activity, Arch. Biochem. & Biophys. **51**: 251, 1954.
18. Ellenbogen, L., Williams, W. L., Rabiner, S. F., and Lichtman, H. C.: Improved Urinary Excretion Test as an Assay for Intrinsic Factor, Proc. Soc. Exper. Biol. & Med. **89**: 357, 1955.
19. Glass, G. B. J., Boyd, L. J., and Luhby, A. L.: Intrinsic Factor Assay: Correlation of Hepatic Uptake of Radioactive Vitamin B_{12} With Hematologic Response in Pernicious Anemia, J. Lab. & Clin. Med. **46**: 60, 1955.
20. Adams, E. B.: Anemia in Kwashiorkor, Brit. M. J. **1**: 537, 1954.

21. Walt, F., Holman, S., and Hendrickse, R. G.: Megaloblastic Anemia of Infancy in Kwashiorkor, and Other Diseases, Brit. M. J. **1**: 1199, 1956.
22. Kho Lien-Keng, Soedjono D. Poesponegoro, and Poey Seng Hin: The Bone Marrow Picture of Kwashiorkor in Djakarta, Doc. Med. Gogr. Trop. **9**: 69, 1957.
22a. Kho Lien-Keng, and Odang, O.: Megaloblastic Anemia in Infancy and Childhood in Djakarta, A. M. A. J. Dis. Child. **97**: 209, 1959.
23. MacIver, J. E., and Back, E. H.: Megaloblastic Anaemia of Infancy and the Anaemia of Malnutrition in Jamaica, West Indian M. J. **7**: 161, 1958.
24. Aldrich, R. A., and Nelson, E. N.: Megaloblastic Anemia in Infants, Lancet **67**: 399, 1947.
25. Faber, H. K.: Value of Liver Extract (343) in Identifying and in Treating Certain Anemias of Infancy and Childhood, Am. J. Dis. Child. **36**: 1121, 1928.
26. Parsons, L. G.: Studies in Anemias of Infancy and Early Childhood, Arch. Dis. Childhood **8**: 85, 1933.
27. Rominger, E., Meyer, H., and Bomskov, C.: Anämiestudien am wachsenden Organismus, über die Pathogenese der Ziegenmilchanamie, Ztschr. ges. exper. Med. **89**: 786, 1933.
28. Bachman, A. L.: Macrocytic Hyperchromic Anemia in Early Infancy, Am. J. Dis. Child. **52**: 633, 1936.
29. May, C. D., Nelson, E. N., Lowe, C. U., and Salmon, R. J.: Pathogenesis of Megaloblastic Anemia in Infancy, Am. J. Dis. Child. **80**: 191, 1950.
30. May, C. D., Stewart, C. T., Hamilton, A., and Salmon, R. J.: Infection as a Cause of Folic Acid Deficiency and Megaloblastic Anemia, A. M. A. Am. J. Dis. Child. **84**: 718, 1952.
31. Prugansky, J., and Axelrod, A. E.: Antibody Production to Diphtheria Toxoid in Vitamin Deficiency States, Proc. Soc. Exper. Biol. & Med. **89**: 323, 1955.

" . . . Hence it is that if the heart be unaffected, life and health may be restored to almost all other parts of the body; but the heart being chilled, or smitten with any serious disease, it seems matter of necessity that the whole animal fabric should suffer and fall into decay. When the source is corrupted, there is nothing, as Aristotle says, which can be of service either to it or aught that depends on it. And hence, by the way, it may perchance be wherefore grief, and love, and envy, and anxiety, and all affections of the mind of a similar kind are accompanied with emaciation and decay, or with cacochemy and crudity, which engender all manner of diseases and consume the body of man. . . ."

William Harvey, M.D., "An Anatomical Disquisition on the Motion of the Heart and Blood in Animals."

From Cardiac Classics, by Frederick A. Willius and Thomas E. Keys

COAGULATION DISORDERS IN THE NEWBORN PERIOD

S. VAN CREVELD, M.D.
NETHERLANDS

PATHOGENESIS

DISTURBANCES which occur in the clotting mechanism of newborn infants can be divided into three groups.

1. Transitory deficiencies in substances necessary for coagulation due to the physiological immaturity of infancy is one type of disturbance. These are often more marked, and persist longer, in premature infants.

2. Another disturbance is congenital abnormalities in the clotting mechanism, which are usually of a permanent nature.

3. Temporary or permanent disturbances in coagulation secondary to an independent condition is the third.

To fully understand the abnormalities that occur in blood coagulation, it is necessary to first briefly review the modern concept of the clotting mechanism. It should be realized that these ideas are continually being modified and that there is currently considerable differences of opinion as to the role of the various coagulation factors. I will outline what I feel is the consensus of opinion among the investigators most active in the field.

The first phase of blood coagulation is concerned with the formation of circulating thromboplastin. At least three plasma factors and one platelet factor are necessary for this. The three plasma thromboplastic factors are the antihemophilic factor (AHF), plasma thromboplastin component (PTC), and plasma thromboplastin antecedent (PTA).

In the second phase, the conversion of prothrombin to thrombin takes place. The thromboplastin formed in the first phase as well as calcium and the so-called prothrombin conversion factors are involved in this process. Various names are given to this last group. The most common are factor VII or proconvertin and factor V or proaccelerin.

In the third phase, fibrinogen is changed to fibrin under the influence of thrombin.

Other factors necessary for coagulation have been recently described, such as Stuart-Prower factor, factor X, and Hageman factor. Of these, probably the most important in newborn infant physiology is the Stuart-Prower factor which is necessary for the formation of thromboplastin as well as the conversion of prothrombin to thrombin.

Transitory Deficiencies in Substances Necessary for Coagulation Due to Organ Immaturity.—Between the second and fifth day after birth, newborn infants have a coagulation defect which usually disappears by the end of the first week of life. The

From the Department of Pediatrics, University of Amsterdam.

bleeding diathesis caused by this abnormality is referred to as hemorrhagic disease of the newborn infant (Townsend[74]) or morbus hemorrhagicus neonatorum (Salomonsen[61]). The most frequent manifestation of the condition is neonatal melena.

The etiology of the hemorrhagic disease of the newborn infant was obscure until 1937. Brinkhous, Smith, and Warner[7] were the first to demonstrate the existence of a decreased prothrombin content in the plasma of full-term newborn infants. The prothrombin levels directly after birth range from 14 to 39 per cent of the normal adult values. After the first day of life, they found that the prothrombin content rose very slowly to reach normal adult values after one year.

With a few exceptions, the findings of Brinkhous and co-workers were confirmed by a large number of investigators. However, most observers found a further decrease of the prothrombin content on the second and third day after birth. Although a number of workers found the prothrombin content to reach normal by about the seventh day of life, the majority agree with the findings of Brinkhous and his colleagues; namely, that adult values are not reached until the end of the first year of life.[13, 27, 55, 59, 76a]

It soon became apparent that the etiology of morbus hemorrhagicus neonatorum was not solely a physiological hypoprothrombinemia. This was suggested by the fact that the addition of stored serum from adults, which contained no fibrinogen, thrombin, or prothrombin, considerably shortened the greatly prolonged prothrombin time of plasma from newborn infants.[60, 70] The factor supplied by the serum was later discovered to be proconvertin. It was then realized that a transitory deficiency in proconvertin was an important factor in the etiology of the hemorrhagic disease of the newborn infant.[12, 27, 45, 50, 52, 76a] It is now felt by some that the apparent decrease in prothrombin activity in newborn infants is due mainly to the deficiency in proconvertin. At the present time, there is great disagreement concerning the actual concentration of prothrombin in full-term infants at birth and later on in infancy.

The prothrombin and proconvertin abnormalities are more pronounced and persist longer in premature than in full-term infants. Premature infants also show marked variation in prothrombin content during the first week of life.[12, 13, 37] The activity of proaccelerin, the other prothrombin conversion factor, in the plasma of full-term and premature infants has also been studied. Although some investigators found decreased values of this factor in the plasma during the first hours after birth, no significant abnormalities could be found after the third day of life. Others found the concentration of proaccelerin to be normal at birth and to remain normal during the first week of life in the plasma of full-term and premature infants.[6, 13, 26, 30, 31, 36, 39, 42, 59, 63, 76a]

There have been a number of studies on the concentration of fibrinogen in the plasma of normal newborn infants. Although most of the investigators found a slight decrease in the fibrinogen concentration during the first days of life, the abnormality

was not felt to be severe enough to interfere with the formation of fibrin. It is therefore felt that hypofibrinogenemia does not play a significant role in the hemorrhagic disease of the newborn infant.[6, 10, 11, 44, 62, 72]

Decreased levels of the Stuart-Prower factor were recently found in the blood of newborn premature infants by Schulz and van Creveld.[68] They found a marked decrease in this factor in a case of hemorrhagic disease of a newborn infant. The recently described serum thromboplastin factor, factor X,[41] should also be decreased in the blood of a newborn infant. However, there is still a question as to the relationship of this factor to PTA and the newly described Stuart-Prower factor. Abnormalities in the fibrinolytic system producing transitory increased fibrinolysis in the plasma of newborn infants have also been described.[53, 54, 63] Using the thromboplastin generation test, a transitory deficiency of plasma thromboplastin component (PTC) was found in newborn, full-term, and premature infants.[1, 14, 15, 28, 30, 31, 36] This abnormality was more marked in the premature infants and was demonstrable for several weeks.[14, 15]

The etiology of the coagulation defect found in newborn infants is felt to be due to a combination of abnormal vitamin K utilization, its inadequate formation and absorption,[24, 37, 51, 77, 78] and immaturity of liver function. The latter observation helps to explain the fluctuation in the various coagulation factors plus the variations in response to vitamin K therapy. Immaturity in liver function also explains reported temporary combined deficiencies in PTA, fibrinogen, and other factors.[1, 16, 36, 43]

Congenital Abnormalities in the Clotting Mechanism.—Congenital deficiencies producing hemorrhagic diseases have been reported of almost all of the factors necessary for coagulation. These may occur individually or in various combinations. Vascular abnormality with or without an associated coagulation defect have also been described. Included in this group are the following: (1) hemophilia A or classic hemophilia or antihemophilic globulin deficiency; (2) hemophilia B or Christmas disease or PTC deficiency; (3) hemophilia C or PTA deficiency; (4) vascular hemophilia or angiohemophilia[18, 67]; (5) vascular PTC deficiency; (6) several forms of thrombopathia, including thrombopathia hemophilica[21a]; (7) congenital idiopathic thrombopenic purpura; (8) congenital idiopathic hypoprothrombinemia[17]; (9) congenital hypoproconvertinemia[19, 20, 23]; (10) parahemophilia (congenital factor V deficiency); (11) congenital shortage of Stuart-Prower factor[3, 4, 38a, 72a]; (12) congenital a- and hypofibrinogenemia; (13) multiple hereditary deficiencies.

The hemorrhagic diatheses produced by the majority of these abnormalities may have their onset in the newborn period. The presence of the abnormality in other members of a family plus the demonstration of the specific coagulation defect aid in establishing the diagnosis in the majority of cases. Occasionally difficulties are encountered in recognizing some of these diseases. This can occur in the milder forms of hemophilia where the coagulation defect may be difficult to demonstrate

and in the sporadic forms of these diseases where the usual family history is not present.

Disturbances in Coagulation Secondary to an Independent Condition Which Can Have Either a Temporary or a Permanent Character.—

The use of drugs by the mother: The use of antiepileptic drugs during pregnancy, just prior to delivery, and during lactation has been implicated as a cause of a hemorrhagic state in newborn infants. In these cases abnormalities in prothrombin, proconvertin, and the Stuart-Prower factor, appearing as late as the fourth day of life, have been reported. These abnormalities as well as the accompanying hemorrhagic symptoms disappeared rapidly following the injection of vitamin K_1 (1 mg.) and of K_3.[22, 68] Phenobarbital was found to accentuate the transitory hypoprothrombinemia of the newborn infant.[25] The other commonly used antiepileptic, Dolantin, was found to occasionally produce hepatic damage with jaundice and the coagulation defects associated with liver disease.

The maternal ingestion of other drugs besides the antiepileptic group has been found to produce coagulation defect in the newborn infant. Such drugs include the coumarin derivatives, salicylates, sulfonamides, and reserpine. Whether or not the coumarin derivatives pass from mother to child during pregnancy and lactation has not been definitely established as yet. Most investigators assume that such a transfer indeed takes place, and some are of the opinion that the influence of the coumarin drugs on the prothrombin level of the baby is even greater than on that of the mother.[32] The risk is felt to be especially great in the beginning of the lactation.[71]

The effect of congenital and acquired hepatic disease: Since the liver is of great importance in the formation of the majority of the coagulation factors, decreased hepatic function is frequently accompanied by deficiencies in many of these substances. This is true for both the congenital parenchymal liver disorders as well as those arising shortly after birth. In mild liver disease, abnormalities in prothrombin and in proconvertin are found; in severe parenchymal liver disease, deviations in proaccelerin, PTC, fibrinogen, and PTA may occur.[13, 49] In the future, the recently discovered Stuart-Prower factor may be implicated in these cases. A deficiency of antihemophilic factor has not been observed in liver disease.

The coagulation disturbances accompanying hemolytic disease of the newborn infant: Prior to exchange transfusion, a decrease in proaccelerin was found by us in a large percentage of infants with hemolytic disease of the newborn period.[8]

Thrombopenia after exchange transfusion: When citrated blood, stored for more than 24 hours, is used in an exchange transfusion, a thrombopenia occurs within the first 24 hours in a large percentage of cases.[41a] This thrombopenia may be accompanied by a severe hemorrhagic state. When fresh heparinized blood is used, the thrombopenia does not occur until several days after the exchange transfusion and is usually not as marked.[9]

Intrauterine melena: This is a rare coagulation defect caused by a decreased concentration of proaccelerin

(factor V). Although the usual hypoprothrombinemia of newborn infants was slightly exaggerated in two of the three patients studied with this condition, the hemorrhagic symptoms were directly proportional to the concentration of proaccelerin. The three cases described were characterized by hemorrhagic liquor amnii and rectal hemorrhage which occurred immediately after birth. One of the babies had slight hemoptysis, but there were no other symptoms or positive physical findings. Two of the infants studied were premature, the other full term. The condition was not altered by the administration of vitamin K; however, the proaccelerin concentration in all three babies eventually increased with a simultaneous disappearance of hemorrhages. It is felt that this syndrome is caused by a temporary disturbance in the formation of factor V by the liver.[13, 73]

Secondary thrombopenia: There are a variety of conditions which may be accompanied by a secondary thrombopenia. These include: (1) congenital leukemia; (2) congenital lues; (3) bacterial or viral infection. (An example of this is the "generalized cytomegalic inclusion body disease of infancy." When this disease occurs in the newborn infant, it can strongly resemble morbus hemolyticus neonatorum and is usually accompanied by purpura caused by thrombopenia); (4) congenital toxoplasmosis with thrombopenia; (5) thrombopenia, accompanied by a decrease of megakaryocytes, with or without generalized bone marrow depression (Schulman[67a]); (6) thrombopenia accompanying erythroblastosis fetalis; (7) thrombopenia in infants of mothers with idiopathic thrombopenic purpura[35, 47, 48]; (8) thrombopenia secondary to maternal ingestion of certain drugs[2]; (9) thrombopenia in infants whose mothers have received frequent blood transfusions; (10) thrombopenia in infants with extensive chronic eczema during the first weeks of life.

DIAGNOSIS

To determine the etiology of a coagulation disorder, it is necessary to ascertain the specific defect responsible for the hemorrhagic state. The principles and techniques of the laboratory procedures employed have been recently described in several textbooks and review articles.[66]

The diagnostic criteria used to establish a deficiency in the newly described Stuart-Prower factor are to be found in other sources.[4, 38a, 68, 72a] Not only have congenital deficiencies of this factor been described but in a recent study low values of the Stuart-Prower factor were found in newborn premature infants.[68] The Stuart-Prower factor is present in normal plasma and serum and is indispensable for both the first and second phase of coagulation. Therefore, a deficiency of this factor results in an abnormal thromboplastin generation test with the patient's serum as well as an abnormal prothrombin time. Since most reagents used to measure proconvertin activity are also lacking adequate Stuart-Prower factor, a decrease in the latter will give falsely low values for proconvertin. However, with a Stuart-Prower deficiency, the Russell viper venom clotting time remains abnormal, but it is normal with a proconvertin deficiency.

As noted previously, the hemorrhagic diseases which occur in the

newborn period can be divided into three groups. In evaluating a newborn infant with a hemorrhagic disorder, initial efforts should be made to determine into which of these three groups the syndrome belongs. One can start by determining which phase of coagulation is disturbed. With exception of the rare case of a- and hypofibrinogenemia, a disturbance of phase 1 will be accompanied by a decrease in prothrombin consumption, while a prolonged prothrombin time will be found in abnormalities of phase 2. More specialized investigations can be attempted to determine the specific defect.

Morbus Hemorrhagicus Neonatorum.—The findings in this entity are as follows: (1) A prolonged prothrombin time which will return to normal after the administration of vitamin K and which is usually due to a simultaneous deficiency in both prothrombin and proconvertin may be present. (2) In premature infants there may be a deficiency in PTC which can be demonstrated by an abnormal thromboplastin generation test (TGT) with the serum of the patient. (3) Occasionally there is a deficiency of fibrinogen and PTA.

Congenital Abnormalities in the Clotting Mechanism.—These may be determined by demonstrating the following specific defect involved in each abnormality.

Hemophilia A (AHG deficiency): (1) prolonged clotting time or prolonged recalcification time (not necessary); (2) an abnormal TGT with plasma of the patient; (3) high residual prothrombin in the serum; (4) low level of antihemophilic factor (especially important for recognizing mild forms of AHF deficiency, i.e., AHF = 5 to 10 per cent); (5) specific correction studies using plasma from patients with known hemophilia A and comparing the results obtained with normal plasma; (6) positive family history with sex-linked transmission (absent in sporadic cases and in rare cases with an abnormal hereditary course).

Hemophilia B (PTC deficiency): (1) high residual prothrombin in the serum; (2) an abnormal TGT with serum from the patient; (3) specific correction studies with serum obtained from a patient with hemophilia B and comparing the results obtained with normal serum; (4) family history.

Hemophilia C (PTA deficiency): (1) abnormal thromboplastin generation test when plasma and serum of the patient are run simultaneously; (2) failure of the patient's plasma to correct a defect of plasma from a patient with PTA deficiency; (3) abnormal thromboplastin generation test which is corrected by substituting either barium-sulfate-treated plasma or serum from a normal individual.

Congenital idiopathic hypoprothrombinemia: (1) prolonged prothrombin time from birth which does not respond to administration of vitamin K; (2) decreased prothrombin content of plasma; (3) family history.

A- or hypofibrinogenemia: (1) may or may not have a positive family history; (2) no coagulation after adding thrombin to the plasma; (3) quantitative determination revealing markedly decreased or absence of fibrinogen.

Parahemophilia (rarely manifests itself in the newborn period): A prolonged prothrombin time which is corrected by the addition of fresh plasma but not by addition of stored plasma from a normal individual. (Several patients previously reported to have hypoproconvertinemia recently appeared to have a deficiency of the newly discovered Stuart-Prower factor.)

Vascular hemophilia: All of the characteristic findings of the classic hemophilia A plus a prolonged bleeding time are present.

Vascular PTC deficiency: All of the characteristic findings of PTC deficiency plus a prolonged bleeding time are present.

Diagnosis of Coagulation Disorders Secondary to Independent Conditions.—Diagnosis of this disorder in the newborn period requires an extensive investigation to determine the basic pathology. In hepatocellular disease and obstructive jaundice, there may be deficiencies of varying degrees in several of the coagulation factors, producing a hemorrhagic state.

TREATMENT

Intelligent therapy of hemorrhagic states in the neonatal period can only be accomplished when the correct diagnosis has been established. Abnormalities in many of the coagulation factors can produce clinical pictures which are very similar. For example, melena neonatorum which is usually caused by a hypoprothrombinemia and hypoproconvertinemia can also be produced by a deficiency in factor V, and may be accompanied by thrombopenia and hypofibrinogenemia.

Hemorrhagic Disease of the Newborn Infant.—In 1936 Dam and associates established the relationship between hypoprothrombinemia and a deficiency of vitamin K in experimental animals. In 1939 several investigators independently came to the conclusion that the physiological hypoprothrombinemia of the newborn infant was due to an inadequate formation and reabsorption of vitamin K during the first days after birth.[24, 37, 51, 77, 78] They showed that the prolonged prothrombin time both of normal newborn infants and of newborn infants with hemorrhagic disease could be prevented by either giving vitamin K to the mother at the beginning of delivery or to the baby immediately after birth. Subsequently, a marked improvement of the PTC deficiency in full-term infants was found following the administration of vitamin K_1.[1] This improvement was not as effective in premature infants.

Recent experience in the treatment of morbus hemorrhagicus neonatorum has caused some reservation about the value of vitamin K in the therapy of this condition. In the first place, the effect of vitamin K on premature infants was found to be far less than on full-term infants. This indicates that a deficiency of vitamin K is not the most important factor in producing the coagulation defect found in the hemorrhagic disease of the newborn infant. Probably more important is the insufficient production of the coagulation factors secondary to immaturity of liver function. In the second place there is considerable difference of opinion concerning the influence of vitamin K on the frequency of hemorrhages in newborn

infants. Nevertheless, vitamin K remains the treatment of choice in cases of hemorrhagic disease of the newborn infant.

Despite the fact that Danish investigators originally pointed out that very small doses of vitamin K were sufficient to forestall the physiological hypoprothrombinemia in newborn infants,[25] far too much vitamin K has been given for many years. The objections which have been made against these high doses and especially against high doses of certain vitamin K preparations are known and shall not be discussed at this time.[21]

We now give 1 mg. of vitamin K_1 to premature infants on the first day of life. In infants with hemorrhagic manifestations, we repeat this dose on the second day either intramuscularly or, when melena exists, as powder via the mouth. In severe hemorrhagic manifestations, transfusions of fresh blood are given when indicated.

Congenital Abnormalities in the Clotting Mechanism Which Are Usually of a Permanent Nature.—For the majority of these conditions, fresh blood and fresh plasma in addition to local therapy with a thrombin preparation are effective. In cases of hemophilia A, stored plasma should not be used because of decreased AHF activity with storage. In newborn infants with hemophilia, the necessity to use an AHF preparation will seldom exist. This is also true of vascular hemophilia. Patients with Christmas disease (PTC deficiency) theoretically can be treated with stored plasma or serum. However, in our experience, better results are obtained with fresh blood or plasma. In PTA deficiency, blood and plasma which do not necessarily have to be fresh may be used.

Transfusions of fresh blood and fresh plasma are required for the treatment of newborn infants with either the rare congenital deficiencies in factor V (parahemophilia) or those secondary to liver disease and sepsis.

In platelet abnormalities such as thrombopathia or congenital thrombopenia, transfusions of fresh blood collected in silicone glass may be effective. Because of the danger of severe infections in newborn infants, splenectomy should not be performed. Since cerebral hemorrhages are infrequent in infants, splenectomy is rarely urgent. Our experiences with prednisone in thrombopenia are not very favorable.

The defect of congenital hypoproconvertinemia can be partially and temporarily corrected by transfusions of serum, plasma, or blood. Congenital a- and hypofibrinogenemia may be treated with blood (plasma) or fibrinogen solutions.

The therapy of coagulation disorders in newborn infants caused by morbid conditions belonging to group C depends on the nature and extent of these disorders.

REFERENCES

1. Aballi, A. J., Banús, V. L., de Lamerens, S., and Rozengvaig, S.: Coagulation Studies in the Newborn Period, A. M. A. J. Dis. Child. **94**: 589, 1957.
2. Ackroyd, J. F.: The Pathogenesis of Thrombocytopenic Purpura Due to Hypersensitivity to Sedormid, Clin. Sc. **7**: 249, 1949.
3. Bachmann, F., Duckert, F., Geiger, M., Baer, P., and Koller, F.: Differentiation of the Factor VII Complex. Studies on the Stuart-Prower Factor, Thromb. Diath. haem. **1**: 169, 1957.

4. Bachmann, F., Duckert, F., and Koller, F.: The Determination of the Stuart-Prower Factor, Thromb. Diath. haem. **2**: 24, 1958.
5. Barkhan, P.: Christmas Factor Activity of Cord-blood, Brit. J. Haemat. **3**: 215, 1957.
6. Beller, F. K.: Die Gerinnungsverhältnisse bei den Schwangeren und beim Neugeborenen, Leipzig, 1957, Johann Ambrosius Barth.
7. Brinkhous, K. M., Smith, N. P., and Warner, E. D.: Plasma Prothrombin Level in Normal Infancy and in Hemorrhagic Disease of the Newborn, Am. J. M. Sc. **193**: 475, 1937.
8. de Bruijne, J. I., and van Creveld, S.: Clotting Factors in Haemolytic Disease of the Newborn, Neo-Natal Studies **4**: 149, 1955.
9. de Bruijne, J. I., van Creveld, S., and Liem Khe Hoo: Clotting Factors in Haemolytic Disease of the Newborn. II. Thrombocytopenia After Replacement Transfusion, Neo-Natal Studies **5**: 109, 1956.
10. Burstein, M., Levi, S., and Walter, P.: Sur l'existence du fibrinogène foetal, Sang **25**: 102, 1954.
11. Crane, M. M., and Sanford, H. M.: Instances Involved in the Coagulation of the Blood of the Newborn Infant; Variations in the Fibrinogen Content in the Normal Infant, Am. J. Dis. Child. **51**: 99, 1936.
12. van Creveld, S., Paulssen, M. M. P., Ens, J. C., van der Meij, C. A. M., Versteeg, P., and Versteegh, E. T. B.: Proconvertin Content in the Blood of Full-term and Premature Infants, Neo-Natal Studies **3**: 53, 1954.
13. van Creveld, S., Paulssen, M. M. P., and Teng, S. K.: Prothrombin and Accelerator Globulin in the Plasma of Newborns Under Normal and Pathological Conditions, Neo-Natal Studies **1**: 87, 1952.
14. van Creveld, S., Nagel, L. G. M., Nijenhuis, J. H., Miranda, S. I., and Tjon Sien Kie, A. P.: Thromboplastin Formation in the Blood of the Newborn, Neo-Natal Studies **3**: 135, 1954.
15. van Creveld, S., Baker, H., Niessing, I., Sypkema, J. J., and Smits, C.: Thromboplastin Formation in the Blood of the Newborn Infant, Neo-Natal Studies **3**: 217, 1954.
16. van Creveld, S., and Mochtar, I. A.: The Transitory Absence of Several Clotting Factors in a Newborn Baby, Arch. Dis. Childhood **29**: 290, 1954.
17. van Creveld, S.: Congenital Idiopathic Hypoprothrombinaemia, Acta paediat. (Suppl. 100) **43**: 245, 1954.
18. van Creveld, S., Jorndan, F. L. J., Punt, K., and Veder, H. A.: Deficiency of Anti-hemophilic Factor in a Woman, Combined With a Disturbance in Vascular Function, Acta med. scandinav. **151**: 381, 1955.
19. van Creveld, S., Veder, H. A., and Blans, M. M.: Congenital Hypoproconvertinaemia, Ann. paediat. **187**: 373, 1956.
20. van Creveld, S., Veder, H. A., and Blans, M. M.: Hypoproconvertinémie Congénitale, Sang **28**: 23, 1957.
21. van Creveld, S.: Vroeggeboorte, Hyperbilirubinemie en kernicterus, Nederl. tijdschr. geneesk. **101**: 705, 1957.
21a. van Creveld, S., and Paulssen, M. M. P.: A Form of Hemorrhagic Diathesis Characterized by the Lack of the Third Clotting Factor Normally Present in Blood-platelets, Ann. paediat. **101**: 193, 1953.
22. van Creveld, S.: Morbus haemorrhagicus neonatorum, Nederl. tijdschr. geneesk. **101**: 2109, 1957.
23. van Creveld, S., and Veder, H. A.: Congenital hypoproconvertinaemia. II, Ann. paediat. **190**: 316, 1958.
24. Dam, H., Tage-Hansen, E., and Plum, P.: Vitamin K Lack in Normal and Sick Infants, Lancet **2**: 1157, 1939.
25. Dam, H., Dyggve, H., Larsen, H., and Plum, P.: The Relation of Vitamin K Deficiency to Hemorrhagic Disease of the Newborn, Advances Pediat. **5**: 129, 1952.
26. Douglas, A. S., and Davies, P.: Hypoprothrombinemia in the Newborn, Arch. Dis. Childhood **20**: 509, 1955.
27. Dyggve, H.: Prothrombin and Proconvertin in the Newborn and During the First Year of Life, Acta paediat. **27**: 251, 1958.
28. McElfresh, A. E., Sharpsteen, J. R., and Akabane, T.: The Generation of Thromboplastin and Levels of Plasma Thromboplastin Component in the Blood of Infants, Pediatrics **17**: 870, 1951.
29. Epstein, R. D., Lozner, E. L., Cobbey, T. S., Jr., and Davidson, C. S.: Congenital Thrombocytopenic Purpura. Purpura Hemorrhagica in Pregnancy and in the Newborn, Am. J. Med. **9**: 44, 1950.
30. Fresh, J. W., Ferguson, J. H., and Lewis, J. H.: Blood-clotting Studies in Parturient Women and the Newborn, Obst. & Gynec. **7**: 117, 1956.
31. Fresh, J. W., Ferguson, J. H., Stamey, C., Morgan, F. M., and Lewis, J. N.: Blood Prothrombin, Proconvertin and Proaccelerin in Normal Infancy: Questionable Relationships to Vitamin K, Pediatrics **13**: 241, 1957.
32. Gordon, R. R., and Dean, T.: Foetal Deaths From Antenatal Anticoagulant Therapy, Brit. M. J. **2**: 719, 1955.
33. Grossman, B. J., Heijn, R. M., and Rozenfeld, J. H.: Coagulation Studies in the Newborn Infant, Pediatrics **9**: 182, 1952.

34. Harrington, W. J., Hollingsworth, J. W., Minnich, V., and Moore, C. V.: Demonstration of a Thrombocytopenic Factor in the Blood of Patients With Idiopathic Thrombocytopenic Purpura, J. Clin. Invest. **30:** 646, 1951.
35. Harrington, W. J., and Sprague, C. C.: Immunologic Mechanisms in Idiopathic and Neonatal Thrombocytopenic Purpura, A. M. A. Arch. Int. Med **38:** 433, 1953.
36. Hartman, J. R., Howell, D. A., and Diamond, L. K.: Disorders of Blood Coagulation During the First Week of Life, A. M. A. Am. J. Dis. Child. **90:** 594, 1955.
37. Hellman, L. M., and Shettles, L. B.: Factors Influencing Plasma Prothrombin in the Newborn Infant, Bull. Johns Hopkins Hosp. **65:** 138, 1939.
38. Heyn, R. M., Rozenfeld, I. H., Grossman, B. J., and Stuart, J. D.: Coagulation Studies in the Newborn Infant. II. Erythroblastosis Fetalis, Pediatrics **9:** 327, 1952.
38a. Hougie, C., Barrow, E. M., and Graham, J. B.: Stuart Clotting Defect. I and II, J. Clin. Invest. **36:** 485 & 497, 1957.
39. Israels, L. J., Zipursky, A., and Sinclair, C.: Factor V Levels in the Neonatal Period, Pediatrics **15:** 180, 1955.
40. Koch, F.: Blutungsübel bei Neugeborenen, Klin. Wchnschr. **34:** 174, 1956.
41. Koller, F.: Factor X, Rev. hémat. **10:** 362, 1955.
41a. Krevans, J. R., and Jackson, D. P.: Hemorrhagic Disorder Following Massive Whole Blood Transfusions, J. A. M. A. **159:** 171, 1955.
42. Larrieu, M. J., Soulier, J. P., and Minkowski, A.: Le sang du cordon ombilical: étude complète de la coagulabilité, comparison avec le sang maternel, Études Néo-Natales **1:** 39, 1952.
43. Lelong, M., Alagille, E. T., and Gentil, C.: Syndrome hémorragique néonatal tardif, lié à l'absence totale et transitoire du facteur anti-hémophilique B, de proconvertine et de prothrombine, Arch. franç. pédiat. **15:** 663, 1958.
44. Lelong, M., Rossier, A., Alagille, E. T., and Marchand, E.: Étude fonctionnelle due foie chez le prématuré a l'aide du complexe prothrombique et de l'életrophorèse, Rev. Intern. Hépatol. **5:** 1130, 1955.
45. Loeliger, A., and Koller, F.: Behaviour of Factor VII and Prothrombin in Late Pregnancy and in the Newborn, Acta haemat. **7:** 157, 1952.
46. Mauer, A. M., Devaux, W., and Lahey, M.: Neonatal and Maternal Thrombocytopenic Purpura Due to Quinine, Pediatrics **19:** 84, 1957.
47. Mendel, E. B., and Sparkman, R.: Idiopathic Thrombocytopenic Purpura in Pregnancy, J. Internat. Coll. Surgeons **28:** 156, 1957 (Abst. Surg. Gynec. & Obst. **106:** 265, 1958).
48. Naeslund, J.: Untersuchungen in den Fibrinogengehalt des Blutes bei Neugeborenen unter Berücksichtigung der Verhältnisse bei Haemophilia neonatorum temporaria, Arch. obst. & gynec. scandinav. **14:** 143, 1934.
49. Naeye, R. L.: Hemophilioid Factors: Acquired Deficiencies in Several Hemorrhagic States, Proc. Soc. Exper. Biol. & Med. **94:** 623, 1957.
50. de Nicola, P.: Factor VII (SPCA): Its Physio Pathologic Significance, Blood **8:** 947, 1953.
51. Nygård, K. K.: Prophylactic and Curative Effect of Vitamin K in Hemorrhagic Disease of the Newborn, Acta obst. & gynec. scandinav. **19:** 361, 1939.
52. Owen, C. A., and Hurn, H. M.: Changes in Blood Coagulation Factors During the First Week of Life, J. PEDIAT. **42:** 42, 1953.
53. Passaro, G.: La fibrinolisi ematica nel neonato, Pediat. internaz. **4:** 247, 1954.
54. Passaro, G., and Grassini, A.: La fibrinolisi ematica nelle emopatie del bambino, Arch. ital. pediat. e puericolt. **17:** 504, 1955.
55. Plum, P.: The Prothrombin Content of the Blood During the First Years of Life, Acta paediat. **38:** 526, 1949.
56. Pritchard, R. W., and Vann, R. L.: Congenital Afibrinogenemia: Report on a Child Without Fibrinogen and Review of the Literature, A. M. A. Am. J. Dis. Child. **88:** 703, 1954.
57. Quick, A. J., and Grossman, A. M.: Concentration of Prothrombin in Blood of Babies (3-7 Days Old), Proc. Soc. Exper. Biol. & Med. **40:** 647, 1939.
58. Quick, A. J., and Grossman, A. M.: Prothrombin Concentration in Newborn, Proc. Soc. Exper. Biol. & Med. **41:** 227, 1939.
59. Quick, A. J., Murat, L. G., and Hussey, C. V.: The Prothrombin of the Baby at Birth, J. Lab. & Clin. Med. **40:** 929, 1952.
59a. Quick, A. J., Murat, L. G., Hussey, C. V., and Burgess, G. F.: The Prothrombin in the Newborn, Surg. Gynec. & Obst. **95:** 671, 1952.
60. Randall, A., and Randall, J. P.: Prothrombin Deficiency in the Newborn, Proc. Soc. Exper. Biol. & Med. **70:** 215, 1949.
61. Saito, M., Gittleman, I. F., Pincus, J. B., and Sobel, A. E.: Plasma Protein Pattern in Premature Infants of Varying Weights on the First Day of Life, Pediatrics **17:** 657, 1956.
62. de Sousa, C. S., Ferreira, J. M. C., Gomes, A. F., and Estrela, A.: La fibrinogénémie du nouveau-né, Arch. franç. pédiat. **10:** 474, 1953.

63. de Sousa, C. S.: Les altérations de la coagulabilité sanguine chez le nouveau-né, Pédiatrie **9:** 787, 1954.
64. Salomonsen, L.: Morbus Haemorrhagicus Neonatorum (Hypoprothrombinemia Neonatorum), Acta paediat. **27:** suppl. 1, 1939.
65. Schoen, E. J., King, A. L., and Duane, R. T.: Neonatal Thrombocytopenic Purpura, Pediatrics **17:** 72, 1956.
66. Schulman, I., and Smith, C. H.: Coagulation Disorders in Infancy and Childhood, Advances Pediat. **9:** 231, 1957.
67. Schulman, I., and Smith, C. H.: Vascular Hemophilia. A Familial Hemorrhagic Disease in Males and Females Characterized by Combined Anti-hemophilic Globulin Deficiency and Vascular Abnormality, A. M. A. Am. J. Dis. Child. **90:** 526, 1955.
67a. Schulman, I., Smith, C. H., and Ando, R. E.: Congenital Thrombopenic Purpura, A. M. A. Am. J. Dis. Child. **88:** 784, 1954.
68. Schulz, J., and van Creveld, S.: (To be published in Neo-Natal Studies).
69. Smith, C. A.: The Newborn Patient, Pediatrics **16:** 254, 1955.
70. Stefanini, M.: Activity of Plasma Labile Factor in Disease, Lancet **1:** 606, 1951.
71. von Sydow, G.: Hypoprothrombinemi och hjärnskada hos barn till dikumarinbehandlad moder, Nord. med. **34:** 1171, 1947.
72. Taylor, P. M.: Concentration of Fibrinogen in the Plasma of the Normal Newborn Infants, Pediatrics **19:** 233, 1957.
72a. Telfer, T. P., Denson, K. W., and Wright, D. R.: A "New" Coagulation Defect, Brit. J. Haemat. **2:** 308, 1956.
73. Teng, S. K.: Thesis, Amsterdam, 1952.
74. Townsend, C. W.: The Haemorrhagic Disease of the Newborn, Arch. Pediat. **11:** 559, 1894.
75. Vandenbroucke, J., and Verstraete, M.: Thrombocytopenia Due to Platelet Agglutinins in the Newborn, Lancet **1:** 593, 1955.
76. Vecchio, F., Schettini, F., and Piomelli, S.: Su una nuova sindrome emorragica in una neonata (deficit associato di PTC, fattore VII e prothrombina), Pediatria **2:** 188, 1956.
76a. Vest, M., and Meier, W.: Ueber das Verhalten des Prothrombins, Proconvertins, Proaccelerins, Fibrinogens und des Heparintoleranztests beim Neugeborenen und beim Säugling im Verlauf des ersten Lebensjahres, Ann. paediat. **189:** 282, 1957.
77. Waddell, W. W., Jr., du Guerry, P. III, Bray, W. E., and Kelley, O. R.: Possible Effects of Vitamin K on Prothrombin and Clotting Time in Newly-born Infants, Proc. Soc. Exper. Biol. & Med. **41:** 181, 1939.
78. Waddell, W. W., Jr., and Lawson, G. M.: Hemorrhagic Diathesis of the Newborn, J. A. M. A. **115:** 1416, 1940.

"The diseases of children are generally regarded as difficult to distinguish, and consequently difficult to treat. I am of opinion that this difficulty is more apparent than real; and to those who will take ordinary pains to acquaint themselves with the peculiarities of diseases in children, their characters will present themselves in very intelligible forms . . . . "

"Whooping Cough: Its Nature and Treatment" by John Armstrong, M.D., Gravesend. From the British Medical Journal, May 14, 1859.

CONGENITAL AND NEONATAL THROMBOCYTOPENIC PURPURA

A Review

Eugene Kaplan, M.D.
Baltimore, Md.

THROMBOCYTOPENIC purpura is a symptom complex of multiple etiology characterized by bleeding into the skin and mucous membranes in association with numerical reduction in circulating platelets. Thrombocytopenia is far more commonly the cause for bleeding in the newborn period than would appear from the literature and must always be considered when hemorrhagic symptoms appear during infancy. This review is concerned with congenital and neonatal forms of thrombocytopenic purpura.

Thrombocytopenia may result from faulty megakaryocyte production or excessive peripheral removal of platelets. Fixed states of marrow hypoplasia and megakaryocyte depletion clearly cause chronic thrombocytopenia. Less certain is the relationship of megakaryocyte alterations[1-3] during infection or immune reactions to acute thrombocytopenia. Platelets are believed to be mechanically trapped within the vascular channels of certain tumors[37] or the chronically enlarged spleen.[1-3] Shortened platelet life span may result from platelet injury by antibodies[12, 13, 19] or by the manipulations and conditions of blood storage and transfusion.[26, 27]

The relationship of thrombocytopenia to disturbances in hemostasis[9, 11] grows more complex with each advance in the laboratory identification of separate platelet factors.[1, 10] At the clinical level many combinations of platelet and bleeding abnormality can be recognized. The bleeding tendency in thrombocytopenia can be entirely absent, present to a mild degree, or develop into a serious hemorrhagic disorder. In addition chronic capillary bleeding in the absence of thrombocytopenia may result from qualitative platelet abnormalities[17, 18] or primary vascular defects.

Bleeding may be spontaneous or follow minor trauma and involve the skin, mucous membranes, or internal viscera. Whereas the amount of blood loss is ordinarily of little consequence, the possibility of serious visceral hemorrhage, particularly involving the brain, is a constant concern. This event may occur at any time, but it is to be expected more when bleeding is generally severe or in the immediate postnatal period following the stresses of the birth process.

Thrombocytopenic purpura is associated with a large variety of circumstances. In the newborn and young infant, this disorder is readily separated into distinct clinical categories, acute and chronic. The acute thrombocytopenic state is self-limited, ordinarily lasting several days to several weeks, and clinically benign except for the hazard of intracranial

From The Departments of Pediatrics, Sinai Hospital of Baltimore, Inc., and Johns Hopkins University School of Medicine.

hemorrhage. The chronic thrombocytopenic state is more likely to be permanent and to be associated with serious underlying disease. A classification based on this clinical distinction is proposed in Table I.

TABLE I. CONGENITAL AND NEONATAL THROMBOCYTOPENIC PURPURAS

A. ACUTE
1. Due to maternal antibodies
With maternal purpura
Maternal I.T.P.
Maternal drug sensitization
Without maternal purpura
Maternal isoimmunization to platelets
Maternal isoimmunization to erythrocytes
2. Transfusion thrombocytopenia
3. Neonatal infection
Bacterial, viral, spirochetal
4. Acute neonatal thrombocytopenia, etiology obscure
B. CHRONIC
1. Megakaryocyte hypoplasia
Congenital hypoplastic thrombocytopenia
Congenital hypoplastic anemia (Fanconi)
Congenital leukemia
Congenital reticuloendotheliosis (Letterer-Siwe)
2. Peripheral platelet depletion
Familial purpura with chronic eczema and otitis media
Giant hemangioendothelioma
Chronic idiopathic thrombocytopenic purpura

ACUTE THROMBOCYTOPENIA

The frequent association of maternal purpura with *transient thrombocytopenia in the newborn infant* has received great emphasis[5-8] and provides a striking example of the role of platelet antibodies in the causation of clinical purpura.[13, 19, 20] Maternal platelet antibodies may be produced during the course of maternal idiopathic thrombocytopenic purpura, as a result of specific drug sensitization or of isoimmunization to platelet antigen derived from the fetus or prior transfusion. These antibodies may cross the placental barrier and cause neonatal thrombocytopenia by reaction with fetal platelets.

In *chronic idiopathic thrombocytopenic purpura* (I.T.P.) *of the mother*, platelet antibodies have been demonstrated in the sera of both mother and thrombocytopenic infant.[1, 13, 19-22] These antibodies persist in the mother despite prior splenectomy which might have caused marked improvement in her clinical disease. The severity of disease in the newborn infant is influenced more by the severity of hemorrhagic manifestations during the pregnancy than by maternal age or parity. In this and other immunothrombocytopenic states of the newborn infant, cutaneous petechiae and ecchymoses usually appear in the first 24 hours. Within a few days there may be additional bleeding, such as epistaxis, melena, hematuria, or blood oozing from the cord, and this tendency may develop or continue for as long as 2 to 3 months. More often, all bleeding subsides within the first 5 to 10 days of life. Platelets increase to normal values after the cessation of bleeding; platelet antibodies may persist for longer periods. It is noteworthy that such neonatal immunothrombocytopenia may occur without any clinical bleeding. There is some serologic evidence that in chronic I.T.P. absence of maternal antibodies coincides with normal offspring,[20] whereas the presence of antibodies results in successive infants with transient thrombocytopenia.

Acute neonatal thrombocytopenia is seen in infants born to *mothers with acute purpura who have been exposed to quinine*.[23, 24] Maternal sensitization to quinine is initiated by prior exposure. The antibody in this system

reacts only in the presence of the offending drug.[12, 16] Thus, re-exposure to quinine may result in acute thrombocytopenic purpura, and placental transfer of drug and specific antibody may result in similar injury to fetal platelets. Laboratory confirmation of this sequence has been provided by the demonstration of quinine reactive platelet antibodies in both maternal and fetal plasma.[24] Both mother and baby developed purpura when quinine had been administered just prior to labor. The platelet antibodies persisted for several months, whereas the purpura and thrombocytopenia lasted only several days. No doubt similar instances of maternal drug sensitization involving other chemicals known to be capable of inducing platelet reactions will be recognized. Important in this large group are the sulfonamides, quinidine, Sedormid, para-aminosalicylic acid, and certain anticonvulsants.[12]

Acute neonatal thrombocytopenia may result from *maternal isoimmunization* which does not affect maternal platelets. Platelet antibodies may be produced in the mother following transfusions or exposure to fetal platelet antigen in pregnancy. These antibodies which are incapable of reaction with her platelets cross the placenta and react with fetal platelets. The true incidence and significance of this immunothrombocytopenia awaits clarification of the present disputed status of platelet groups and antigens.[14, 15] It has been described in one family with 3 consecutive purpuric infants[25] and in another family with involvement of one of 3 infants.[1]

Thrombocytopenic purpura may accompany *severe erythroblastosis fetalis* and has occasioned repeated search for platelet antibodies in maternal and fetal serum. With a single notable exception,[1] these attempts have failed, and again this clearly indicates the present contradictions in platelet serology.

Not to be confused with the purpura of severe erythroblastosis which often is present at birth and precedes replacement transfusion is the occasional purpura and thrombocytopenia which is caused by *replacement transfusion*.[26, 27] Platelets in stored bank blood, unless collected and administered under very special conditions, have a greatly shortened survival. This is particularly true if the storage exceeds 24 hours. During exchange transfusion, these defective platelets replace those of the infant and thrombocytopenia may result.

Acute thrombocytopenic purpura may appear at the onset or in the course of severe *neonatal infections*. These include bacterial sepsis and suppurative foci, generalized viral infections such as herpes viremia and cytomegalic inclusion disease, and congenital syphilis. The mechanisms responsible for thrombocytopenia during infection are in dispute, although presumptive evidence favors a disturbance in platelet survival rather than in platelet production. Bleeding is often limited to minor cutaneous and mucosal hemorrhage but may be aggravated by simultaneous hypoprothrombinemia of hepatic failure. This group of purpuras is usually differentiated without difficulty by the clinical manifestations of severe neonatal infection, such as pallor, icterus, hepatosplenomegaly, and generalized toxicity.

At this time it is still impossible to account for the majority of the acute

thrombocytopenias of early infancy.[6, 28] Relatively few are proved to be associated with specific infection, maternal purpura, or platelet antibody. This failure reflects, among other factors, the uncertain techniques of platelet serology and of pediatric virology. The potential frequency and obscure etiology of this syndrome in the newborn infant is well illustrated by a study of acute thrombocytopenia in 75 infants observed during 18 months in one premature station.[29] Maternal purpura could not be implicated, antibodies could not be demonstrated in random specimens of maternal or infant sera, and megakaryocytes appeared adequate in several infants subjected to marrow aspiration. In a large group, thrombocytopenia coincided with mild rectal bleeding after the first week of life. An equivalent number of premature infants with similar rectal bleeding were not thrombocytopenic. In a small group, acute thrombocytopenia appeared in the first days of life; some remained asymptomatic, several died with intracranial hemorrhage, while most developed a mild bleeding disorder with cutaneous and mucosal hemorrhages. A final group developed transient thrombocytopenia without bleeding during the course of mild postnatal infections.

CHRONIC THROMBOCYTOPENIA

Chronic thrombocytopenic purpura in early infancy contrasts sharply with the more common acute thrombocytopenic states in etiology and prognosis. The underlying disturbance is generally genetically determined, the hemorrhagic manifestations are more refractory to therapy, and intracranial bleeding is more likely to lead to fatal outcome. Chronic thrombocytopenia in this age group is the result of persistent megakaryocyte hypoplasia or disturbances which remove platelets at an excessive rate.

Congenital hypoplastic thrombocytopenia[1, 28, 30-33] is a rare familial syndrome characterized by complete or marked depletion in megakaryocytes without other marrow abnormality. It is usually associated with other congenital anomalies, most prominent of which is congenital absence of both radii. Cardiovascular and urogenital anomalies have also been reported as well as congenital spherocytic hemolytic anemia. The relationship of this syndrome to congenital hypoplastic anemia (Fanconi) and congenital leukemia is not at all clear. In its severe form, hypoplastic thrombocytopenia is characterized by an early bleeding tendency which is severe and generalized. Most infants succumb to fatal intracranial hemorrhage within the first months of life. Milder forms of this disease which have remissions of variable duration and which follow a brief period of bleeding in infancy have been described. These remissions, accompanied by some increase in platelets, have occurred spontaneously or in response to the use of platelet transfusion, steroids, or splenectomy. Indeed, the condition may be compatible with prolonged survival in the absence of further clinical hemorrhage, despite continued megakaryocyte depletion and thrombocytopenia.

Chronic thrombocytopenia in association with megakaryocyte hypoplasia is also to be found in the more generalized marrow disturbance of *congenital hypoplastic anemia, congenital leukemia,* and certain cases of *Letterer-Siwe disease.* The clinical course and prognosis in these conditions reflect

the underlying disease process as well as the effects of thrombocytopenia on hemostasis.

Many cases of *giant hemangioendothelioma*[34-38] in infancy are complicated by chronic thrombocytopenia. It is believed that platelets are mechanically sequestered within the vascular channels of this tumor; thrombocytopenia characteristically appears as the tumor grows and disappears as the tumor size regresses. The purpuric manifestations are frequently mild and intermittent, although serious intracranial and visceral hemorrhage have been encountered. Eventual complete recovery following spontaneous or postradiation tumor disappearance has been the outcome in the majority of affected infants.

Chronic eczema and otitis media[39-41] may be associated with chronic thrombocytopenic purpura in infancy. The familial distribution of this unusual syndrome has suggested a sex-linked genetic transmission. The mechanism for thrombocytopenia is not clear since megakaryocytes are not reduced and splenomegaly is characteristically absent. Studies of platelet antibodies and survival are not available. Food allergy may be implicated to some degree. A persistent bleeding tendency begins early in life, most commonly expressed as a petechial rash or as mild melena. The hemostatic response to steroids and splenectomy has been disappointing. Death from overwhelming infection occurred in 5 children in one series, in 4 of whom splenectomy had been performed.[41] In another family the disease has been essentially mild in several children and compatible with long survival without complication.[40]

Chronic idiopathic thrombocytopenic purpura is a disease seen often in adults, infrequently in childhood, and rarely in early infancy. It may begin as early as the first month of life and continue for many years. Remissions and exacerbations in platelet levels and bleeding manifestations characterize the course, and steroid therapy and splenectomy can be at least temporarily beneficial. The cause for this disturbance is uncertain, but it would appear to be affected by platelet survival more than by megakaryocyte production.

DIAGNOSIS

Recognition of reduction in the platelet count is the key to the *laboratory diagnosis* of thrombocytopenic purpura. Platelet numbers in the newborn infant do not differ significantly from those in older subjects and may range between 150,000 to 350,000 per cubic millimeter.[3, 4] The precise choice of method among the many available for the enumeration of platelets is less important than the care used by an experienced technician in performing the test. Indeed, there is often an exaggerated fear to attempt platelet evaluation except under the most ideal conditions. Simply stained peripheral blood films can be made with reasonable care and reveal vital presumptive information to the interested pediatrician. Thus, if platelets appear plentiful and obviously well distributed, thrombocytopenia is immediately excluded. If platelets appear reduced, with some care taken to examine all portions of the smear for gross platelet aggregates, then more expert technical procedures are required for proper confirmation of thrombocytopenia. Counts of less than

100,000 per cubic millimeter represent definite thrombocytopenia.

Prolongation of bleeding time and increased capillary fragility are classical laboratory abnormalities found in thrombocytopenia, although not limited to this condition. Relatively simple to perform in the small infant, the estimation of bleeding time and vascular fragility is especially important since these tests provide a direct measurement of capillary hemostatic function. In general terms, the degree of abnormality in these three laboratory findings correlates with each other and with the clinical severity of the bleeding tendency. Unfortunately, this correlation is unreliable in many instances of thrombocytopenic purpura. Marked reduction of platelet count to 10 to 50,000 per cubic millimeter may coincide with active bleeding on one patient, yet in another no hemorrhagic disturbance will be observed. In a surprising number of infants with acute thrombocytopenia and mild purpura, bleeding time, capillary fragility, or both have been normal.

Diminished plasma prothrombin activity and delayed whole blood coagulation are not characteristic of thrombocytopenia. In the newborn period, diminished plasma prothrombin activity accompanies a physiologic and transitory deficiency in prothrombin and stable factor. Hemorrhagic disease of the newborn infant due to such a coagulation factor deficiency simulates the bleeding of thrombocytopenia but is usually circumscribed between the second and fifth days of life. Congenital coagulation factor deficiencies, single or multiple, may cause persistent abnormalities in prothrombin activity or coagulation time and continuation of hemorrhagic symptoms. Precise diagnosis then requires more elaborate hematologic investigation.[1-4, 9, 10]

Additional laboratory tests of blood coagulation are of lesser clinical value. Reduction in the platelet content of blood is characteristically associated with poor clot retraction and diminished prothrombin utilization. Measurement of thromboplastin generation is a difficult laboratory procedure of more academic than practical import in thrombocytopenia and is pertinent to specific functional abnormalities of platelets.

Bone marrow examination is of limited value in acute thombocytopenic states but assumes major importance when marrow hypoplasia, congenital leukemia, or reticuloendotheliosis are under consideration. It needs to be emphasized that a single marrow aspirate which suggests inadequate megakaryocytes is not a definitive measure of megakaryocyte distribution. Whereas plentiful megakaryocytes in films of marrow aspirate are of certain significance, inadequate numbers may be more apparent than real and should indicate repeated marrow study and perhaps definitive marrow biopsy. This is at least as true in the small infant, where aspiration may prove technically difficult, as in the older child.

Serologic methods for the identification of platelet groups and antibodies are still unsuited for the general clinical laboratory. The diagnosis of immunothrombocytopenia in the newborn infant can be presumed in the presence of maternal purpura. The definitive exclusion or recognition of

platelet antibodies will then depend on highly specialized laboratory procedures.[1, 12, 13, 15, 20]

MANAGEMENT AND TREATMENT

The proper *management of congenital and neonatal thrombocytopenic purpura* can be considered only against the background of the wide etiologic and clinical spectrum just described. It must be firmly emphasized that in the majority of infants thrombocytopenia is of brief duration, entirely free of hemorrhage, or associated with mild and self-limited bleeding manifestations. These infants require no special treatment other than optimal pediatric care. When hemorrhage is more persistent or when clinical signs suggest early intracranial involvement (convulsions, tremors, etc.), specific therapeutic measures should be instituted.[1, 42, 43] These measures include the transfusion of platelets or their products, the use of ACTH or the steroid hormones, and splenectomy.

Whereas ordinary blood transfusion is entirely without effect on the bleeding manifestations of thrombocytopenia, the transfusion of special platelet preparations[43-45] is often dramatically successful. Such preparations include platelet-rich blood from polycythemic donors, platelet concentrates from normal donors, platelet extracts[46, 47] and viable platelets in freshly drawn blood exposed only to silicone-lined or plastic surfaces.[30] Marked improvement in bleeding may begin within an hour of such replacement therapy and last for several days. The hemostatic effect exceeds in extent and duration of improvement in the platelet count and other laboratory findings. However, platelet transfusions have a serious limitation. Their repeated use in any patient results in the development of platelet antibodies by the recipient, with progressive diminution of their benefit. This therapy should be restricted to the management of the acute hemorrhagic crisis or to the preparation of a bleeding patient for surgery. It is particularly adapted to the needs of the newborn infant with acute thrombocytopenic purpura in whom intracranial hemorrhage is suspected and deserves more widespread use of platelet transfusion in this specific situation.

ACTH and adrenal steroids have distinct beneficial effects on the bleeding disorder of many thrombocytopenic states.[1, 42, 48] These effects are complex and involve at least several mechanisms. Most immediate and desirable is a nonspecific improvement in vascular fragility with a decrease in hemorrhagic activity. Days later platelets may significantly increase, whether as a result of inhibition of platelet removal or stimulation of platelet production. For management of the acute hemorrhagic crisis in the small infant, these agents have the virtues of availability, safety, and rapid response. In most chronic thrombocytopenic states of infancy, however, they are entirely without effect or effective partially and for limited periods of time. The recommended daily dosage for ACTH is 10 to 30 U., for cortisone 25 to 50 mg., and for prednisone 10 to 30 mg.

Until recent years, splenectomy was the treatment of choice for the severe hemorrhagic manifestations of thrombocytopenic purpura.[49] When effective, hemostasis is dramatically and quickly improved, and platelets increase within a short time thereafter. In the chronic thrombocytopenic states, this effect is

less constantly observed and of variable duration. The introduction of hormonal and platelet replacement therapy has greatly limited the need for emergency splenectomy. The real threat of postsplenectomy sepsis and meningitis[41, 50, 51] has further dampened pediatric enthusiasm for this procedure. Splenectomy should be reserved as a last resort in the acute hemorrhagic crisis of the young infant, after medical measures have proved ineffective. It has a limited role in the chronic thrombocytopenic states of infancy, where it may be tried in an attempt to stimulate megakaryocyte production or to decrease platelet depletion. The influence of maternal hemorrhagic activity in chronic I.T.P. on the severity of fetal disease has led to the suggestion that splenectomy be performed prior to pregnancy or in its early months. Continued steroid therapy during pregnancy may also be indicated in the presence of severe maternal disease.[1, 42]

The transmission of platelet antibodies from mother to infant via breast milk is unproved and is unlikely to be of harmful effect in view of the proved safety of mother's milk in Rh erythroblastosis. The transmission of a drug like quinine from a sensitized mother is more likely to be of significance and might warrant prohibition of nursing.

SUMMARY

Thrombocytopenia should always be considered in the clinical evaluation of hemorrhage in the newborn and young infant. Often recognizable with no more difficulty than the careful perusal of a peripheral blood film, the presence of low platelets should initiate efforts to establish the responsible mechanism. The onset and duration of bleeding and the presence or lack of accompanying illness or anomalies contribute direction to these efforts. In the newborn period, mild thrombocytopenic purpura in an infant without other disturbances suggests immunothrombocytopenia and the passive transferral of maternal antibody. This suspicion may be supported by deliberate examination of the mother with emphasis on inspection for purpura, blood examination for thrombocytopenia, and taking a careful history for evidence of a bleeding tendency, recent suspicious drug exposure, or previous births resulting in transient neonatal purpura.

Persistence of thrombocytopenia beyond the immediate neonatal period might still reflect a maternal influence. More likely, however, is the possibility of primary illness in the infant, whether congenital or acquired. The presence of other congenital anomalies suggests underlying megakaryocyte hypoplasia, whereas the presence of signs of illness, such as anemia, icterus, visceromegaly, skin rash, and failure to thrive, might indicate one of many serious disease states, such as infection, leukemia, or reticuloendotheliosis.

Management of the thrombocytopenic state in a newborn infant requires judicious watching since the disorder is so frequently benign and self-limited. The attending physician bears considerable responsibility, however, in estimating the imminence of intracranial hemorrhage and in initiating proper therapeutic measures. When thrombocytopenia is chronic, the management is directed at the underlying cause, if possible, and toward attempts to prevent serious bleeding by supporting vascular hemostasis, when necessary, with steroid hormones, splenectomy, or platelet transfusions.

REFERENCES

General Discussion and Reviews of Neonatal Purpura

1. Stefanini, M., and Dameshek, W.: The Hemorrhagic Disorders: A Clinical and Therapeutic Approach, New York, 1955, Grune & Stratton, Inc.
2. Quick, A. J.: Hemorrhagic Diseases, Philadelphia, 1957, Lea & Febiger.
3. Wintrobe, M. W.: Clinical Hematology, ed. 4, Philadelphia, 1956, Lea & Febiger.
4. Wolman, I. J.: Laboratory Applications in Clinical Pediatrics, New York, 1957, McGraw-Hill Book Co., Inc.
5. Stening, S. E. L.: Congenital or Neonatal Thrombocytopenia, M. J. Australia **40:** 210, 1953.
6. Robson, H., and Walker, C.: Congenital and Neonatal Thrombocytopenic Purpura, Arch. Dis. Childhood **26:** 126, 1951.
7. Epstein, R. D., Lozner, E. L., Cobbey, T., and Davidson, C. S.: Congenital Thrombocytopenic Purpura; Purpura Hemorrhagica in Pregnancy and the Newborn, Am. J. Med. **9:** 44, 1950.
8. McAlenney, P., and Kristan, J.: Thrombocytopenic Purpura in the Newborn, Am. J. Dis. Child. **78:** 401, 1949.

Platelet Physiology, Serology Methods

9. Biggs, R., and MacFarland, R. G.: Human Blood Coagulation and Its Disorders, Springfield, 1953, Charles C Thomas.
10. Tocantins, L. M.: The Coagulation of Blood, Methods of Study, New York, 1955, Grune & Stratton, Inc.
11. Ackroyd, J.: Role of Platelets in Coagulation, Thrombosis and Haemostasis, British M. Bull. **11:** 21, 1955.
12. Ackroyd, J.: Platelet Agglutinins and Lysins in Thrombocytopenic Purpura, British M. Bull. **11:** 28, 1955.
13. Tullis, J.: Identification and Significance of Platelet Antibodies, New England J. Med. **255:** 541, 1956.
14. Blattner, R. J.: Platelet Groups and Types, J. PEDIAT. **47:** 382, 1955.
15. Gurevitch, J., and Nelken, D.: ABO Groups in Blood Platelets, J. Lab. & Clin. Med. **44:** 562, 1954.
16. Shulman, N. R.: Immunoreactions Involving Platelets, J. Exper. Med. **107:** 711, 1958.
17. Braunsteimer, H., and Pakesch, F.: Thrombocytoasthenia and Thrombocytopathia—Old Names and New Diseases, Blood **11:** 965, 1956.
18. McIlvanie, S. K.: Thrombopathic States, J. A. M. A. **106:** 2114, 1958.

Immunothrombocytopenia

19. Harrington, W. J., Sprague, C. C., Minnich, V., Moore, C. V., Ahlvin, R. C., and Dubach, R.: Immunologic Mechanisms in Idiopathic and Neonatal Thrombocytopenic Purpura, Ann. Int. Med. **38:** 433, 1953.
20. Harrington, W. J., Minnich, V., and Arimura, G.: The Autoimmune Thrombocytopenias, *in* Progress in Hematology, vol. 1, New York, 1956, Grune & Stratton, Inc.
21. Schoen, E. J., King, A. L., and Duane, A. B.: Neonatal Thrombocytopenic Purpura, Pediatrics **17:** 72, 1956.
22. Vandenbroucke, J., and Verstraete, M.: Thrombocytopenia Due to Platelet Agglutinins in the Newborn, Lancet **1:** 593, 1955.
23. Posner, A. C.: Purpura Hemorrhagica Complicating Puerperium, Am. J. Obst. & Gynec. **34:** 155, 1937.
24. Mauer, A. M., DeVaux, W., and Lahey, M. E.: Neonatal and Maternal Thrombocytopenic Purpura Due to Quinine, Pediatrics **19:** 84, 1956.
25. Schulman, I., Smith, C. H., and Ando, R. E.: Congenital Thrombocytopenic Purpura: Observations on Three Infants Born of a Nonaffected Mother; Demonstration of Platelet Agglutinins and Evidence for Platelet Isoimmunization, A. M. A. Am. J. Dis. Child. **88:** 785, 1954.

Thrombocytopenia After Replacement Transfusion

26. Krevans, J., and Jackson, D.: Hemorrhagic Disorder Following Massive Whole Blood Transfusion, J. A. M. A. **159:** 3, 1955.
27. DeBruigne, J. I., van Creveld, S., and Liem Khe Hoo: Thrombocytopenia After Replacement Transfusion, Études Néo-Natales **5:** 109, 1956.

Acute Neonatal Thrombocytopenia, Etiology Obscure

28. Akerren, Y., and Reinand, T.: Thrombocytopenia in the Newborn Period, Acta med. scandinav. Suppl. **246:** 281, 1950.
29. Kaplan, E.: Unpublished Observations.

Congenital Hypoplastic Thrombocytopenia

30. Bell, A. D., Mold, J. W., and Shaw, S.: Study of Transfused Platelets in a Case of Congenital Hypoplastic Thrombocytopenia, British M. J. **2:** 692, 1956.
31. Gross, H., Groh, C., and Weippl, G.: Kongenitale hypoplastische Thrombopenie mit Radiosaplasie, Neue Osterr. Ztschr. f. Kinderheilk. **1:** 574, 1956.
32. Emery, J. L., Gordon, R. R., Rendle-Short, J., Varadi, S., and Warrack, A. J.: Congenital Amegakaryocytic Thrombocytopenia With Congenital Deformities and a Leukemoid Blood Picture in the Newborn, Blood **12:** 567, 1957.
33. Zetterstrom, R., and Strindberg, B.: Sporadic Congenital Spherocytosis Associated With Congenital Hypoplastic Thrombocytopenia and Malformations, Acta paediat. **47:** 14, 1958.

Giant Hemangioendothelioma

34. Weissman, J., and Tagnon, H. J.: Syndrome of Hemangioma and Thrombocytopenic Purpura, A. M. A. Arch. Int. Med. **92:** 523, 1953.
35. Franklin, A. W., and Williamson, D. A. J.: Haemangio-endothelioma With Haemorrhage and Thrombocytopenia, Arch. Dis. Childhood **28:** 490, 1953.
36. Meeks, E. A., Jay, J. B., and Heaton, L. D.: Thrombocytopenic Purpura Occurring With Large Hemangioma, A. M. A. Am. J. Dis. Child. **90:** 349, 1955.
37. Good, T. A., Carnazzo, S. F., and Good, R. A.: Thrombocytopenia and Giant Hemangioma in Infants, A. M. A. Am. J. Dis. Child. **90:** 260, 1955.
38. Scherz, R., Louro, J., and Geppert, L.: Giant Hemangioendothelioma With Associated Thrombocytopenia, J. PEDIAT. **52:** 212, 1958.

Familial Purpura, Eczema, Otitis Media

39. Aldrich, R. A., Steinberg, A. G., and Campbell, D. C.: Pedigree Demonstrating a Sex-linked Recessive Condition Characterized by Draining Ears, Eczematoid Dermatitis and Bloody Diarrhea, Pediatrics **13:** 133, 1954.
40. Wolff, J. A., and Bertricio, M.: A Sex-linked Genetic Syndrome in a Negro Family Manifested by Thrombocytopenia, Eczema, Blood Diarrhea, Recurrent Infection, Anemia and Epistaxis, A. M. A. J. Dis. Child. **93:** 74, 1957.
41. Huntley, C. C., and Dees, S. C.: Eczema Associated With Thrombocytopenic Purpura and Purulent Otitis Media, Pediatrics **19:** 351, 1957.

Treatment

42. Harrington, W. J.: Therapy of the Purpuras, J. Chron. Dis. **6:** 365, 1957.
43. Stefanini, M.: Management of Thrombocytopenic States, A. M. A. Arch. Int. Med. **95:** 543, 1955.
44. McGovern, J. J.: Platelet Transfusions in Pediatrics, New England J. Med. **256:** 922, 1957.
45. van Creveld, S., Paulssen, M. M. P., and Bartels, H. L. J. M.: Transfusions of Suspensions of Blood Platelets in Thrombocytopenia and Thrombopathia, J. Clin. Path. **6:** 41, 1953.
46. Klein, E., Farber, S., Djerassi, I., Toch, R., Freeman, G., and Arnold, P.: The Preparation and Clinical Administration of Lyophilized Platelet Material to Children With Acute Leukemia and Aplastic Anemia, J. PEDIAT. **49:** 517, 1956.
47. Djerassi, I., Klein, E., Farber, S., and Pechet, G.: Correction of Vascular Fragility by a Water Soluble Platelet Fraction in Patients With Secondary Thrombocytopenia, Fed. Proc. **17:** 435, 1958.
48. Dameshek, W., Rubio, F., Mahoney, J., Reeves, W. H., and Burgin, L.: Treatment of Idiopathic Thrombocytopenic Purpura With Prednisone, J. A. M. A. **166:** 1805, 1958.
49. Bluestone, S. S., and Maslow, H. L.: Essential Thrombocytopenic Purpura in the Newborn Infant. Report of First Case Treated by Splenectomy, Pediatrics **4:** 620, 1949.
50. Gofstein, R., and Gellis, S. S.: Splenectomy in Infancy and Childhood, A. M. A. J. Dis. Child. **91:** 566, 1956.
51. Smith, C. H., Erlandson, M., Schulman, I., and Stern, G.: Hazard of Severe Infections in Splenectomized Infants and Children, Am. J. Med. **22:** 390, 1957.

"... If you do remember,
I send it through the rivers of your blood,
Even to the court, the heart, to the seat o' the brain:
And through the cranks and offices of man,
The strongest nerves and small inferior veins
From me receive that natural competency
Whereby they live."

William Shakespeare:
Act I, Scene I, Coriolanus.

From Cardiac Classics, by Frederick A. Willius and Thomas E. Keys

CONSIDERATIONS OF IRON METABOLISM IN EARLY INFANCY

NATHAN J. SMITH, M.D.
MADISON, WIS.

THE occurrence of clinically apparent iron-deficiency anemia is uncommon in infants less than 6 months of age. The recognized instances are limited to prematurely born infants, to infants who have lost significant amounts of blood very early in life, and to infants who are the products of multiple births.[1] Recent advances in the understanding of the metabolism of iron suggest that derangements in the supply of iron to the fetus and the newborn infant may contribute to the high incidence of "nutritional" anemias in infants older than 6 months of age. Likewise derangements in protein metabolism[2] with compromised production of hemoglobin, resembling that accompanying iron-deficiency states, have been recently studied in very young infants. This condition, called "transient dysproteinemia," has prompted renewed interest in the metabolism of iron in the fetus and in the very young infant.

This brief review will summarize the current status of our knowledge of the normal iron demands of the fetus and infant, how these demands may be altered, and how they are satisfied most effectively. Such a discussion can serve to alert the physician to the instances in which supplemental iron is indicated and contraindicated. In addition the deficiencies in knowledge of iron metabolism and the need for further critical investigation in this area will be apparent.

From the Department of Pediatrics, University of Wisconsin School of Medicine.

IRON METABOLISM IN THE FETUS

During normal intrauterine development, the fetus will demand approximately 300 mg. of iron.[3] The total iron content and body weight increase proportionately during gestation, the concentration of iron per unit of fetal weight remaining relatively constant throughout fetal development.[4] Studies of fetal weight and iron content have permitted the calculation of fetal iron demand at various periods of pregnancy. The demand is negligible during the first and second trimesters, being no more than a total of 8.0 mg. In the last trimester the rapidly growing fetus requires approximately 4.0 mg. per day, a total of approximately 300 mg. During the last half of pregnancy, the maternal hemoglobin mass will increase likewise to a degree demanding up to 500 mg. of iron.[5] Thus, it is not surprising that iron-deficiency anemia is a common finding late in pregnancy even in the primipara who has eaten a so-called "normal" diet. The combination of iron contained in blood lost during delivery, iron contained in the placenta, and the 300 mg. of iron required by the infant totals the amount of maternal iron loss during a pregnancy. This loss exceeds by more than 200 mg. the amount of iron conserved by the absence of

menses during pregnancy (250 to 300 mg.). This deficit may well be the basis of a more deficient iron supply available to the fetus during future pregnancies.[6]

The mechanism involved in the transfer of iron across the placenta to the fetus is not known. It has been demonstrated that iron is transported to the placental site in the maternal circulation bound to a beta-1 globulin, transferrin. A similar protein in the fetal circulation is involved in the transport of iron from the placenta to fetal tissues. The concentration of transferrin (iron binding protein) has been found to be significantly higher in the maternal plasma than in the fetal plasma taken from the umbilical vessels. In addition the plasma iron level is distinctly higher in cord blood than in the maternal circulation.[7] It is therefore apparent that the transfer of iron across the placenta to the fetus is against a distinct gradient; the transfer of iron requiring the removal of iron from relatively low concentrations in the maternal plasma to the higher concentrations in the fetal circulation. That this is a one-way transfer mechanism has been demonstrated by the injection of Fe^{59} into the umbilical circulation. Following such an injection, radioactive iron failed to appear in the maternal circulation.[8]

The dynamic processes involved in the cellular transport of iron across the placenta have not been defined. The rate of transfer is extremely rapid, occurring in a matter of a few minutes, and would appear to be governed principally by the fetal need for iron.[10] It can no longer be attributed to the destruction of maternal erythrocytes in the placenta. Wöhler[9] has suggested that the transfer of iron across the placenta is accomplished through the formation of ferritin, having demonstrated that placental ferritin becomes labeled with Fe^{59} from the maternal circulation. The mechanisms involved in this incorporation of maternal iron in the placenta and its subsequent release to the fetus are not known.

As previously mentioned, the fetal demand for iron late in pregnancy usually totals 3 to 4 mg. daily. The maternal iron "turnover" in the process of normal hemoglobin production is between 25 and 30 mg. daily. Thus, the fraction of the total iron "turnover" which goes to the fetus is relatively small. Nevertheless, the amount of iron destined for the fetus may be significantly influenced by factors which alter the maternal "turnover" of iron. Pribilla[11] has shown that abscess formation in animals decreases the maternal iron turnover and likewise diminishes the amount of iron transferred to the fetus. When large amounts of iron were injected into these animals, iron was increased in fetal as well as maternal tissues. The need for extended observations of this kind is apparent as well as a clearer understanding of the mechanisms of placental transfer of iron. It is not unreasonable to postulate that a variety of factors which influence the metabolism of iron in the mother may distinctly alter the transfer of iron to the fetus. Whether or not iron deficiency in the mother interferes with the relatively small amount of iron normally destined each day for transfer to the fetus cannot be answered from direct observation. A detailed study of infants born of iron-deficient mothers would suggest that this might be the case.

THE INFLUENCE OF MATERNAL IRON DEFICIENCY

The relationship of iron-deficiency anemia in the mother to the hemoglobin levels of the newborn infant has been a subject of considerable study Strauss[12] concluded that anemia during pregnancy would lead to anemia during the first year of life in the infant. Several years later Woodruff and Bridgeforth[13] could not confirm any correlation between deficits in hemoglobin in mothers and altered hemoglobin concentrations in their infants. The absorption of iron has been studied as an index of iron "demand" in mothers and infants. No correlation could be established by this method between the mothers' hemoglobin at the time of birth and the amount of iron absorbed by their infants.[14] However, two independent studies have recently demonstrated that infants of low birth weight and infants of high birth order are more likely to develop clinically apparent iron-deficiency anemia during early life. It is postulated that in these situations the amount of iron received from maternal sources was less than normal and that the supply of maternal iron influenced hemoglobin synthesis during infancy.[15, 16]

Difficulties in specifically identifying iron deficiency in pregnant women and quantitating hemoglobin production in very young infants have been in a large part responsible for the inability to satisfactorily define the relationship between hemoglobin production in the infant and iron deficiency in their mothers. Recently Lund[17] has done much to clarify this relationship by using suitable biochemical techniques to determine the presence of iron deficiency and by determining total body hemoglobin as a measure of hemoglobin production. Using the total hemoglobin mass, the levels of free erythrocyte protoporphyrin, and, to a lesser extent, the serum iron levels as indicators of iron-deficiency anemia, these workers have found from 10 per cent to over 50 per cent of pregnant patients to be anemic. In a subsequent study of 66 pregnant women and their infants, it was found that infants born of anemic mothers had a reduction of 20 per cent of the total circulating hemoglobin mass when it was measured after these infants reached 4 to 5 days of age. The more severe the anemia in the mother the more profound the change in the newborn infant. Inasmuch as the principal site of "iron storage" in the newborn infant is that iron in the circulating hemoglobin, these low levels of total hemoglobin can logically be expected to influence the production of iron-deficiency anemia later in infancy.[18]

In an additional series of 38 infants Sisson and Lund[18] attempted to evaluate the effect of early and late interruption of the umbilical circulation at the time of birth on blood volume and hemoglobin mass. Demarsh and coworkers[20] and Duckman and his colleagues[21] studied the so-called "placental transfusion" effect of delayed ligation of the cord and concluded that as much as 60 c.c. of blood and its iron-containing hemoglobin could be salvaged for the infant whose cord was tied late. It is of interest that in these recent studies neither early nor late clamping of the cord with the infant held above or below the level of the placenta significantly altered the total blood volume or hemoglobin mass as measured on the fourth day of life. If the cord were manually milked, higher

hemoglobin contents and plethora were obtained. The results of such studies give satisfying support to the practice of "moderation" in the hitherto confused issue of when to interrupt the cord circulation.

IRON REQUIREMENT IN EARLY INFANCY

The iron requirement of any given infant during the first 6 months of life can be thought of as the difference between the total body iron present at birth and that present in hemoglobin and other body tissues at 6 months of age. Physiologic losses and the excretion of iron is thought to be too minute to be of significance in any consideration of iron needs. The iron requirement is determined principally by the total body iron present at birth and the rate of growth. Under circumstances considered "normal," these two factors may be responsible for as much as a 300 per cent variation in the demand for exogenous iron during the first year of life.[3]

The iron demand in a normal infant can be quite simply calculated as follows: One can begin with a "normal" infant whose birth weight is 3.5 kilograms and whose total blood volume is 85 c.c. per kilogram, with a hemoglobin concentration at birth of 17 Gm. per cent. In each gram of hemoglobin there is 3.4 mg. of iron, which accounts for 173.0 mg. of total iron in the infant under consideration. Such an infant could have 6 mg. per kilogram of tissue iron and estimated iron stores of 35 mg. of iron. Thus, the total body iron present at birth would be 229 mg. If such an infant enjoyed an expected growth experience and weighed 8.0 kilograms at 6 months of age, his normal total body iron would have increased to approximately 300 mg. The 70 mg. iron demand would have been met by assimilation of iron from exogenous dietary sources.

It is of interest to calculate iron demands in this fashion for infants whose iron content might be influenced by commonly encountered variables experienced in early infancy. Infants born of iron-deficient mothers in the group studied by Sisson and Lund[18] had a 20 per cent reduction in their hemoglobin mass at birth. This would represent an iron deficit of approximately 35 mg. in the hypothetical infant weighing 3.5 kilograms, considered above, and an increase of 50 per cent in the total demand for iron in the first 6 months of life. Any loss of infant blood perinatally or during early infancy would likewise add to this demand. A hemorrhage of as little as 20.0 ml. of blood in the newborn period would add approximately 12.0 mg. to the iron requirement.

Rates of growth in excess of that used in the above calculation will obviously be reflected in increases in the iron requirement. This is encountered most strikingly in the prematurely born child. Calculated iron requirements in excess of 110.0 mg. for the first 6 months of life are not uncommon in the premature infant.[22] A less striking increase in iron demand associated with increases in growth rate has recently been suggested by Woodruff[15] in comparing iron demands of male and female infants. A preponderance of male infants was found in a large group of patients suffering from iron-deficiency anemia. The hemoglobin concentration of male infants has been shown to be less than in the female, yet the growth rate of males is greater in the first year of life and their hemoglobin increment is

greater. Therefore, there are greater iron requirements. It is suggested that sex differences in iron metabolism might exist in infancy as they do in adults.

Is it reasonable to assume that "normal" requirements for iron can be satisfied from dietary sources alone? Calculations based on the iron content of foods given in a conventional diet with the introduction of solid foods at 3 months of age indicate an average intake of iron of approximately 5.5 mg. daily. A total of somewhat more than 1.0 Gm. of iron would be ingested during the first 6 months of life. Eighty per cent of this iron is derived from iron-enriched infant cereals. Most of the remaining 20 per cent of the dietary iron is presented in the form of vegetables, fruit, meat, and eggs. The small amount of iron in milk makes an insignificant contribution (78 mg.) to the total iron intake in the first 6 months of life.[22]

That such dietary iron can be assimilated from various dietary sources has been demonstrated recently. Quantitative studies have been made of the absorption of iron from infant foods labeled in vivo with radioactive iron. From 10 to 20 per cent of the iron in milk, cereal, eggs, and liver was absorbed by infants under 6 months of age.

Thus, if an infant ingests 1,000 mg. of iron in an optimal diet in the first 6 months and absorbs an average of 10 per cent of the dietary iron, his calculated demands of 70 mg. should be readily available to him from his diet. It must be emphasized that this situation presupposes an optimal diet which is particularly well supplied with iron-supplemented infant cereal, a normal growth rate, a normal complement of iron stores at birth, and the absence during this period of pathological states such as gastroenteritis, hemorrhage, or infection.

The high incidence of iron-deficiency anemia in pregnant women in the United States has been mentioned. A variety of situations in which fetal and infant blood is lost have recently been described.[23] These situations are not uncommon, and the amount of blood lost by the infant need not be great to significantly disturb the balance in iron demand and supply in the first 6 months of life. Infections, particularly those involving the gastrointestinal tract, are not uncommon in infancy. Even in the face of increased iron demand, i.e., iron deficiency, infants with gastroenteritis are unable to assimilate iron from their diets.[22]

Such studies indicate that there exists a small margin between the demands for iron and that amount available from a normal diet. Likewise there is a frequent occurrence of abnormal situations in very early life in which the demand for iron becomes greater than can be satisfied by the diet. Faced with this situation, consideration has been given to the desirability of supplementing the normal dietary intake of iron of all infants with some form of medicinal iron. Sturgeon[3] has administered iron intramuscularly to a group of "normal" infants. He evaluated resultant changes in hemoglobin levels, iron binding capacity of the serum, free erythrocyte protoporphyrin, serum iron, and serum copper concentrations. Comparison of the results with those found in the study of adequate controls demonstrated that many of the chemical manifestations of apparent "iron deficiency" in normal infancy

can be altered by the administration of intramuscular iron. In studies of the absorption of iron greater absorption was noted in "normal" infants under 1 year of age, being comparable to absorption found in iron-deficiency states in older infants or children.[22] This finding can be interpreted to indicate the presence of an increased need for iron in the early age group.

These studies have been interpreted by some as further evidence demanding the administration of medicinal iron preparations to all infants. There is little doubt that the recent knowledge of the metabolism of iron has clearly demonstrated the need for iron supplementation in a variety of situations where this need had been overlooked in the past. However, to attempt to satisfy the needs for iron in these infants by the "routine" administration of iron to all infants would appear to be an unwarranted departure from critical and thoughtful pediatric practices. A more desirable approach to the assurance of optimal iron nutrition in infancy can be the physician's thoughtful, individual evaluation of the iron needs of the very rapidly growing infant, the infant who has bled, or who has suffered from gastrointestinal disturbances.

ADMINISTRATION OF IRON IN INFANCY

When administration of medicinal iron in infancy is limited to those infants whose prenatal or perinatal experience has resulted in the presence of a deficit in the body's iron content, full therapeutic doses of iron should be given. Indications for the use of lesser doses have not been defined in infancy. Under 6 months of age, adequate doses of orally administered iron salts are in the range of from 45 to 60 mg. of elemental iron. This iron is most readily administered as any one of several "drop" preparations now available. In such preparations iron is usually present in a concentration of 25.0 mg. per cubic centimeter. Doses of from 15 to 30 mg. of elemental iron (0.6 to 1.2 c.c.) can be given 3 to 4 times daily with expectation of optimal assimilation. Smaller doses will have to be given too frequently throughout the day to be practical. When larger amounts are given in a single dose, the percentage of the iron absorbed is reduced.[24] Oral iron medication is best given in a fasting state, one to two hours prior to feedings to avoid the formation of insoluble and unabsorbable iron complexes with phosphates and other constituents of milk. Gastrointestinal intolerance to oral iron is an extremely uncommon occurrence in early life. It will usually be necessary to maintain administration of iron medication for several weeks to three to four months if deficits are to be corrected and adequate replenishment of tissue iron components is to be realized.

A variety of additives have been combined with iron in an attempt to enhance its effectiveness. Molybdenum, copper, vitamin C, cobalt, and vitamin B_{12} are available in a variety of so-called "hematinics" in combination with iron. None of these additives are known to increase the effectiveness of the administered iron, and some of these materials are known to be toxic.[23] When iron is needed, the most effective, least expensive, and safest medication to use is a simple iron salt.

An iron-dextran compound suitable for intramuscular administration is

available. Well-controlled studies indicate that the response to this medication is no more rapid and no greater than that to properly administered oral iron. Intramuscular iron is expensive, potentially toxic, and produces some local discomfort. In light of these considerations the use of this material is appropriately limited to those infants with gastrointestinal disease who cannot tolerate orally administered iron salts.

DISTURBANCES IN IRON AND PROTEIN METABOLISM IN EARLY INFANCY

Considerable interest has been evidenced recently in the metabolic interrelationships of iron and plasma proteins in the early months of life. In 1956 Ulstrom, Smith, and Heimlich[2] reported their experience with four infants less than 7 months of age who had edema, pallor, and a striking degree of irritability. Laboratory study revealed the presence of severe hypochromic microcytic anemia, hypoproteinemia, hypocupremia, and hypoferremia. No other evidences of renal or hepatic disease were present. The hypoproteinemia was characterized by a uniform reduction of the concentration of albumin and of all of the globulin components. Normal growth rates had been experienced by all of the infants. Prenatal and postnatal abnormalities could not be found. Of considerable importance was the fact that in each instance these patients had been taking an entirely "normal" diet. Balance studies indicated that the over-all nitrogen balance was positive, but a greater than expected excretion of urinary nitrogen was demonstrated. Plasma protein turnover rates were determined in three of the infants by using tracer doses of phenylalanine-2-C^{14}. The incorporation of the isotope in each protein fraction was similar in the hypoproteinemic infants and in the controls. However, the isotope disappeared from the circulation at an increased rate in the infants with low plasma proteins. It was postulated that this increased rate of plasma protein degradation without compensatory increases in the rate of protein synthesis was responsible for the hypoproteinemia in these patients. The hypoferremia, hypocupremia, and anemia were secondary to a primary abnormality in plasma protein metabolism. In spite of a high intake of dietary iron in these infants, it was not until medicinal iron was given that the anemia was corrected. Recovery from the hypoproteinemia occurred spontaneously in each patient before 9 months of age, unrelated to any changes in diet or administration of any medication, including iron. The hypocupremia and hypoferremia disappeared as the plasma protein concentration returned toward normal.

At the same time Lahey and Shubert[23] and Sturgeon and Brubaker[26] reported their experiences with similar infants manifesting hypoproteinemia, hypocupremia, hypoferremia, and anemia. The onset of disease in both these groups of patients was somewhat later than in those studied by Ulstrom and co-workers.[2] Of considerable interest was the fact that both Lahey's and Sturgeon's patients were receiving cow's milk as their sole source of food intake. Because of this fact, both of these groups of workers postulated that the condition was the result of a "new" deficiency state, a conclusion not in keeping with the dietary intake of the four infants reported by Ulstrom. The resulting question to be

answered is whether the patients were suffering from one and the same disorder or whether two or more similar clinical states exist with varying etiology.

Cartwright has recently reported an experience with hypocupremic, hypoproteinemic infants and concludes that at least two similar clinical states with differing causes can be recognized in this group of anemic and edematous infants. One group of patients who suffered from a primary plasma protein deficit was similar to those reported by Ulstrom, and perhaps a larger group had a nutritional deficiency as the primary etiologic factor. Lahey as well as Sturgeon are in sympathy with this suggestion. However, until more definitive studies of plasma protein metabolism are available in affected infants, until the clinical findings can be corrected by some specific nutrient, and until a deficit in the body content of some nutrient or its metabolic products is demonstrated, the question of one or more than one etiologic factor must remain open.

That iron and protein metabolism are closely related is shown in areas other than the syndrome of transient dysproteinemia. In approximately 15 per cent of infants with nutritional iron-deficiency anemia, detectable abnormalities in plasma proteins are present. Whether these changes, most commonly hypoalbuminemia, are related to comprised rates of synthesis or rapid loss of the albumin from the circulation remains to be determined. Doubtless the future must provide more precise information as to the relationships of iron and protein metabolism in the early months of life when the demands of growth of these two body constituents are great.

REFERENCES

1. Smith, N. J., and Rosello, S.: Iron Deficiency in Infancy and Childhood, Am. J. Clin. Nutrition **1:** 275, 1953.
2. Ulstrom, R. A., Smith, N. J., and Heimlich, E. M.: Transient Dysproteinemia in Infants, A New Syndrome. I. Clinical Studies, A. M. A. J. Dis. Child. **92:** 219, 1956. II. Studies of Protein Metabolism Using Amino Acid Isotopes, **93:** 536, 1957.
3. Sturgeon, P.: Iron Metabolism. A Review, Pediatrics **18:** 267, 1956.
4. Widdowson, E. M., and Spray, C. M.: Chemical Development in Utero, Arch. Dis. Childhood **26:** 205, 1951.
5. Pribilla, W., Bothwell, T. H., and Finch, C. A.: Iron Transport to the Fetus in Man, *in* Iron in Clinical Medicine, University of California Press, 1958, pp. 58-64.
6. Moore, C. V.: The Importance of Nutritional Factors in the Pathogenesis of Iron Deficiency Anemia, Am. J. Clin. Nutrition **3:** 1, 1955.
7. Smith, C. H., Schulman, I., and Morganthau, J. E.: Iron Metabolism in Infants and Children. Serum Iron and Iron Binding Protein, Diagnostic and Therapeutic Implications, Advances Pediat. **5:** 195, 1952.
8. Bothwell, T. H., Pribilla, W., and Finch, C. A.: Transplacental Iron Transport to the Rabbit (To Be Published).
9. Wöhler, F.: Zur Physiologie und Pathologie des Speichereisens; über den intermediären Eisenstoffwechsel der Plazenta, Deutsche med. Wchnschr. **80:** 30, 1955.
10. Vosberg, G. J., and Flexner, L. B.: Maternal Plasma as a Source of Iron for the Fetal Guinea Pig, Am. J. Physiol. **161:** 202, 1950.
11. Pribilla, W.: Tierexperimentelle Untersuchungen über den Eisenaustausch zweschen Mutter und Foet nach intravenöser Eisengabe, Acta haemat. **12:** 371, 1954.
12. Strauss, M. B.: Anemia of Infancy From Maternal Iron Deficiency in Pregnancy, J. Clin. Invest. **12:** 345, 1933.
13. Woodruff, C. A., and Bridgeforth, E. B.: Relationship Between the Hemogram of the Infant and That of the Mother During Pregnancy, Pediatrics **12:** 681, 1953.
14. Oettinger, L., Jr., Mills, W. B., and Hahn, P. F.: Iron Absorption in Premature and Full-term Infants, J. PEDIAT. **45:** 302, 1954.
15. Woodruff, C. A.: Multiple Causes of Iron Deficiency in Infants, J. A. M. A. **167:** 715, 1958.
16. Guest, G. W., and Brown, E. W.: Erythrocytes and Hemoglobin of the Blood in Infancy and Childhood. III. Factors in Variability, A. M. A. J. Dis. Child. **93:** 486, 1957.

17. Lund, C. J.: The Iron Deficiency Anemia of Pregnancy, Am. J. Obst. & Gynec. **62:** 947, 1951.

18. Sisson, T. R. C., and Lund, C. J.: The Influence of Maternal Iron Deficiency on the Newborn, Am. J. Clin. Nutrition **6:** 377, 1958.

19. Whipple, G. A., Sisson, T. R. C., and Lund, C. J.: Delayed Ligation of the Umbilical Cord, Its Influence on the Blood Volume of the Newborn, Obst. & Gynec. **10:** 603, 1957.

20. Demarsh, Q. B., Windle, W. F., and Alt, H. L.: Blood Volume of the Newborn Infant in Relation to Early and Late Clamping of the Umbilical Cord, Am. J. Dis. Child. **63:** 1123, 1942.

21. Duckman, S., Merk, H., Lehman, W. X., and Regan, E.: The Importance of Gravity in Delayed Ligation of the Umbilical Cord, Am. J. Obst. & Gynec. **66:** 1214, 1953.

22. Schulz, J., and Smith, N. J.: Quantitative Study of the Absorption of Food Iron in Infants and Children, A. M. A. J. Dis. Child. **95:** 109, 1958.

23. Smith, N. J.: Iron as a Therapeutic Agent in Pediatrics, J. PEDIAT. **53:** 37, 1958.

24. Schulz, J., and Smith, N. J.: Quantitative Study of the Absorption of Iron Salts in Infants and Children, A. M. A. J. Dis. Child. **95:** 120, 1958.

25. Lahey, M. E., and Shubert, W. K.: A New Deficiency Syndrome Occurring in Infancy, A. M. A. J. Dis. Child. **93:** 31, 1957.

26. Sturgeon, P., and Brubaker, C.: Copper Deficiency in Infants, A. M. A. J. Dis. Child. **92:** 254, 1956.

27. Zipursky, A., Dempsey, H., Markowitz, H., Cartwright, G., Wintrobe, M. M.: Studies on Copper Metabolism. XXIV. Hypocupremia in Infancy, A. M. A. J. Dis. Child. **96:** 148, 1958.

" . . . we are able to show, by some remarkable experiments, . . . that molecular and tailed forms of some material substance may be seen issuing out of (the corpuscles) and passing into the plasma, without much alteration of their form or colour; . . . there can be no difficulty in concluding that the corpuscles, in their natural state, discharge matter into the plasma. It must be confessed that but little can be demonstrated of the vital and depurating processes constantly going forward in the blood of the living person. Nevertheless, in the absence of such demonstration, we are warranted, by the experiment just referred to, and by the undisputed teaching of physiology, in concluding that the corpuscles of blood are sustained in their vital and chemical qualities by the plasma and the air; also, that their excretions are passed, partly into the plasma and partly into the air . . . . "

Galstonian Lectures on "Fever and Inflammation," delivered before the Royal College of Physicians, London, by William Addison, M.D.

From the British Medical Journal, April 30, 1859.

THE ANEMIA OF PREMATURITY

IRVING SCHULMAN, M.D.
CHICAGO, ILL.

THE fact that prematurely born infants regularly develop anemia is generally recognized. Accumulated hematologic studies have demonstrated, however, that the anemia of prematurity is by no means a single entity and that the premature infant is likely to manifest significant anemia (i.e., hemoglobin concentrations below 10 Gm. per 100 ml.) in three distinct phases during the first year of postnatal life. The characteristic course of the hemoglobin concentration in a premature infant is shown in Fig. 1. It may be noted that the hemoglobin concentration begins to fall soon after birth and reaches a minimum at approximately 7 weeks of age. This is designated as the *early anemia* of prematurity. There follows a period, approximately equal in duration to the early phase, in which the hemoglobin concentration is maintained at the previous low level or rises but slightly. This is the *intermediate phase*. A second fall in hemoglobin concentration then occurs, often resulting in levels even lower than those in the early phase. This is the *late phase* which, if untreated, may persist into the second year of life.

The division of the hematologic course of the premature infant into the aforementioned phases is not arbitrarily done, but is based upon distinct differences in pathogenesis. Fig. 1 demonstrates that the physician may find the same hemoglobin concentration over a period of months in an individual baby. This same level, however, if obtained in each phase, has quite different implications in regard to prognosis and therapy. While each phase evolves from the events in the preceding one, each is based upon completely different underlying mechanisms. The rational management of the anemias of prematurity requires an understanding of the hematologic mechanisms operating during each phase.

From The Children's Memorial Hospital and Northwestern University Medical School.

EARLY ANEMIA OF PREMATURITY

During the first week of postnatal life, the hemoglobin concentration of the premature infant usually rises slightly. Subsequently, however, the hemoglobin concentration falls steadily for 5 to 8 weeks and then rather abruptly levels off. During this period, the red cell count falls proportionately so that a *normochromic anemia* develops. In general the smaller babies demonstrate a more rapid fall in hemoglobin concentration and reach lower hemoglobin levels than do the larger infants. The minimum hemoglobin level averages about 8.0 Gm. per 100 ml. for babies with birth weights of 1,000 to 1,500 grams, 8.5 Gm. per 100 ml. for infants of 1,500 to 2,000 grams, and 9.5 Gm. per 100 ml. for infants of 2,000 to 2,500 grams. In comparing infants in

various weight groups, however, there is considerable overlap in both the rate of fall and minimum hemoglobin concentration, and only a rather poor correlation between birth weight and severity of anemia may be obtained.

There is general agreement that the fall in hemoglobin concentration during the early phase results from a steady decrease in total circulating hemoglobin mass (Fig. 2). Two mechanisms are involved in the loss of hemoglobin mass: (1) a marked decrease in erythropoietic activity beginning shortly after birth,[1-3] and (2) a decrease in the life span of the erythrocytes.[1, 4-7] While the implication of both mechanisms is generally accepted, the relative influence of each has not been completely defined. That there is a rather striking decrease in hematopoietic activity is evidenced by the fall in reticulocyte count[1, 5] and the decrease in marrow erythroid content.[2] In addition the concept that the red cell life span of newborn infants is shorter than normal has received increasing support from recent studies. Schulman and Smith,[1] on the basis of serial changes in circulating red cell volume, estimated the life span of the erythrocytes in premature infants to range from 77 to 98 days (as compared with the normal of 100 to 120 days in children and adults). Metzroth,[4] calculating the life span from reticulocyte ripening time in premature infants, arrived at a figure of 84 days. Mollison[5] measured the life span of placental blood transfused into four anemia premature infants who received simultaneous transfusions of adult blood. Red cell life span, as measured by differential agglutination, was slightly reduced in the placental blood when compared with adult blood in the same recipient. Recently, Hollingsworth[6] by using chromium-51-tagged placental red cells injected into adult recipients demonstrated a definite decrease in erythrocyte life span. Even more significant, Gilardi and Miescher[7] reinjected chromium-51-tagged newborn erythrocytes back into the infants' own circulations and confirmed the decrease in red cell life span.

The early anemia of prematurity is in the majority of infants a self-limited phase in which spontaneous improvement is to be anticipated.[1-3] This results from a return of intensive erythropoietic activity which leads to a rise in total circulating hemoglobin mass. The hemoglobin synthesized at this time is almost entirely of the adult variety.[1] The termination of the early phase is manifested by a sharp reticulocytosis and a leveling off of the hemoglobin concentration despite continuing weight gain[1, 3] (Fig. 2). The return of intensive hematopoietic activity tends to occur earlier in the smaller babies than in the larger, a fact which argues strongly against the idea that the decrease in erythropoietic activity and its subsequent return are functions of maturation.[3]

It is now well established that control of erythropoiesis is mediated through a humoral erythropoietic factor released from one or more centers (pituitary, kidney) under the influence of anemia, hypoxia, and other stimuli.[8] The available evidence suggests that a similar mechanism is operative in the newborn period and is involved in the early anemia of prematurity and the physiologic anemia of the full-term infant. Rooth and Sjöstedt[9] have demonstrated that exposure of newborn rats to continuous

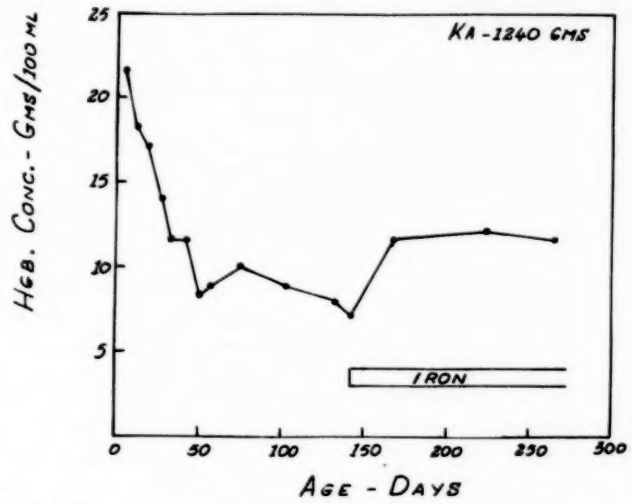


Fig. 1.—Typical course of the hemoglobin concentration in a premature infant.

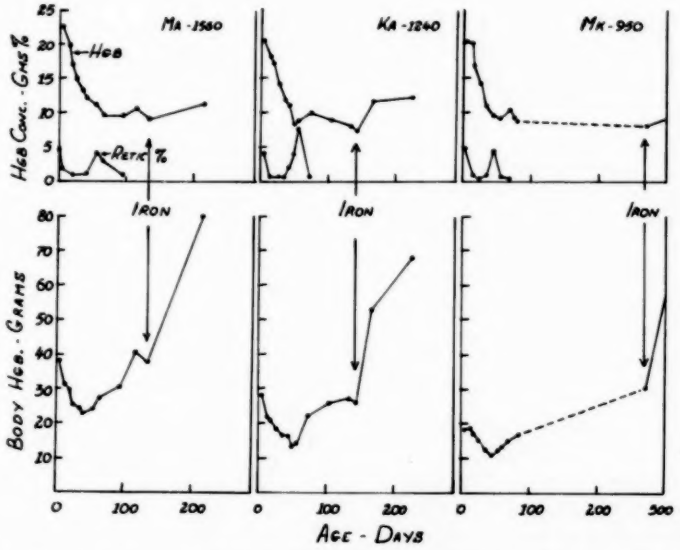


Fig. 2.—Changes in hemoglobin concentration, circulating hemoglobin mass, and reticulocyte percentage in premature infants of different birth weights.

hypoxia after birth results in stimulation of erythropoiesis. In human newborn infants evidences of a similar mechanism may be found. Thus, the baby with cyanotic congenital heart disease continues to exhibit active erythropoiesis and does not develop the so-called physiologic anemia. In addition babies who are born anemic as a result of hemorrhage respond immediately with increased erythropoietic activity. Finally, Seip[10] has demonstrated that premature infants respond to periods of asphyxia with an immediate reticulocytosis of the same magnitude as seen in adults subjected to experimental hypoxia. Based upon these concepts, the decrease in hematopoietic activity in the early weeks of postnatal life would be considered to result from a reduction in the erythropoietic stimulating factor as a consequence of the high hemoglobin concentrations. The return of active erythropoiesis would follow release of the erythropoietic factor as a consequence of the development of anemia.

Gairdner and associates[11] concluded that a hemoglobin concentration of 11 to 12 Gm. per 100 ml. would result in stimulation of erythropoietic activity in the infant. The meticulous reticulocyte studies of Seip and Halvorsen[3] have indicated that this phenomenon may occur at somewhat higher hemoglobin levels.

It would follow from the above considerations that the infant whose hemoglobin concentration falls most rapidly would demonstrate a return of erythropoietic activity earlier than the infant whose hemoglobin concentration falls more slowly. The apparently paradoxical situation wherein the premature infant recovers erythropoietic activity earlier than the full-term infant, and wherein the smaller premature infant demonstrates active regeneration sooner than the larger baby, has been amply documented.[1-3]

An important, and often overlooked, factor which influences all phases of the anemia of prematurity is the hemoglobin concentration and hemoglobin mass with which the premature infant begins life.[12] In a study of 89 premature infants with birth weights less than 1,800 grams, Allen[13] found that the capillary hemoglobin concentrations during the first three days of life varied from 13.8 to 26.0 Gm. per 100 ml. Similar wide ranges of hemoglobin concentration are found throughout gestation and at term.[14] The factors which may influence the hemoglobin concentration during gestation and at birth, such as intrauterine hypoxia, placental bleeding, time of cord clamping, have recently been reviewed[14] and are beyond the scope of this paper. It is evident, however, that differences in starting hemoglobin concentration will greatly affect the hemoglobin levels in the postnatal weeks, the time at which the minimum hemoglobin concentration is reached and the time at which active erythropoiesis returns. Thus, the initial hemoglobin level may significantly affect both the severity and duration of the early phase of the anemia of prematurity. The influence of the hemoglobin concentration at birth and the subsequent course of the early anemia is shown in Fig. 3.

In this study the two infants compared differ only in the starting hemoglobin concentrations, the birth weights and subsequent rates of weight gain being similar. It may be seen that the

infant with the lower initial hemoglobin level continues to manifest a lower hemoglobin concentration during the first 4 weeks of life. However, the minimum hemoglobin concentration and the recovery phase are reached earlier in this infant, so that from the sixth to the tenth week he now has a higher hemoglobin concentration than the baby who had the initial advantage. The hemoglobin concentration in the latter infant continues to progressive loss of circulating hemoglobin mass. Since iron is not excreted from the body, it follows that the early anemia is accompanied by a progressive accumulation of iron stores. This has been documented by studies of both liver and marrow iron.[3, 15] Both organs show virtually no iron at birth and a progressive accumulation of iron in the postnatal weeks. In the marrow the largest amounts of iron are found between

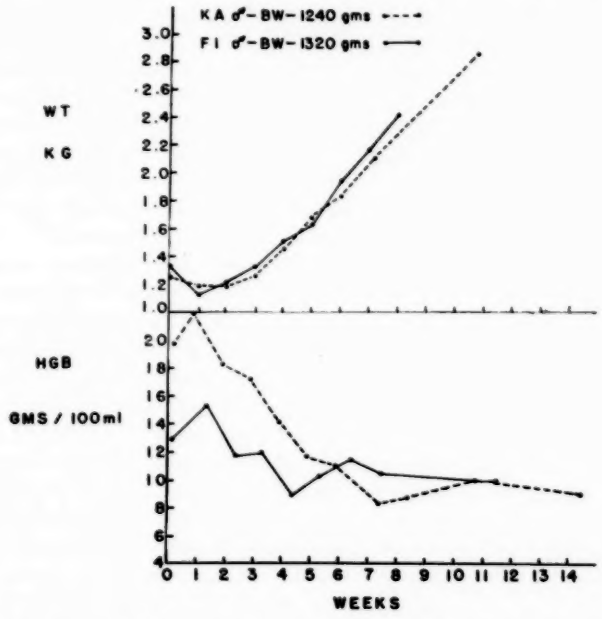


Fig. 3.—The course of hemoglobin concentration in two infants with similar weight curves but with different initial hemoglobin concentrations.

fall for 7½ weeks, reaches an identical minimum level, and then enters the recovery phase. At 3 months of life, the hemoglobin concentrations are equal. These changes speak strongly for the concepts of regulation expressed above.

As mentioned earlier, the early anemia of prematurity results from a the fourth to the eighth week.[3] Thus, the early anemia of prematurity occurs in the face of *iron excess* and the administration of iron in the early weeks of postnatal life would not be expected to modify this phase. This has been confirmed by many studies.[12, 16-18] It should be emphasized that virtually the entire iron store of the premature

infant at birth resides in the circulating hemoglobin mass. It is evident, therefore, that the hemoglobin mass with which the infant begins life may greatly influence the later need for, and response to, supplementary iron. This will be discussed later.

Since the early anemia of prematurity is self limited, transfusions are rarely indicated. It should be stressed, however, that termination of the early phase depends upon the ability of the marrow to respond to the stimulus for increased erythropoiesis. In the presence of clinical situations which are known to retard hematopoiesis, such as infection, the anticipated return of erythropoietic activity may be delayed and transfusions may be required. A great deal of information may be obtained from study of reticulocyte counts in such instances, but careful evaluation remains the best guide. Transfusion solely on the basis of an arbitrarily designated hemoglobin level cannot be recommended.

INTERMEDIATE PHASE

The resumption of active erythropoiesis results in a progressive increase in total circulating hemoglobin mass (Fig. 2). Since weight gain and progressive expansion of blood volume continue throughout this period, a significant increase in peripheral hemoglobin concentration does not occur. During this period, therefore, the hemoglobin concentration remains level or rises only slightly. The progressive increase in hemoglobin mass involves reutilization of the iron which has been liberated and stored during the early phase. The duration of the intermediate phase is therefore directly determined by the considerations of the early phase. Thus, the rapidly growing infant will tend to resume hematopoiesis earlier and will reutilize his iron reserve sooner than the infant growing more slowly. The infant who has a large initial hemoglobin mass will resume hematopoiesis later, will build a larger store of iron, and will manifest a longer intermediate phase than the infant with a low initial hemoglobin level. In general the intermediate phase is approximately equal in duration to the early phase and terminates with the exhaustion of the available iron reserves. Studies of marrow hemosiderin by Seip and Halvorsen[3] have demonstrated a progressive decrease in stainable iron following maximum deposition at the fourth to eighth weeks of postnatal life. In the smallest premature infants the marrows were found to be hemosiderin free at 6 to 8 weeks, in the heavier infants at 8 to 12 weeks.

In 1954 Schulman and Smith[1] concluded that on the basis of available iron stores supplementary iron would not be required for 12 to 16 weeks. Subsequently, the variables of initial hemoglobin mass and rate of weight gain were evaluated and, on the basis of the considerations described above, it became apparent that some babies would require supplementary iron considerably earlier. The most important single factor seemed to be the hemoglobin mass at birth.[12] Although the stored iron is apparently completely reutilized, the question of whether exogenously administered iron might be used preferentially or be more efficiently used has been raised. If this were true, the rate of hemoglobin regeneration in the intermediate phase might be more rapid. Sisson and Whalen[18] concluded that the

initial hemoglobin mass was regained more rapidly in infants receiving iron supplementation (as meat feedings) from 2 to 4 weeks of age than in untreated infants. In the former group the return was considered to occur at 8 weeks as compared to 12 weeks in the latter. It was suggested that "a more adequate reservoir of iron than is naturally available from previous red cell degeneration is needed." However, if one studies the serial changes in individual infants in the study, as opposed to the grouped data, it is apparent that there is considerable overlap between the treated and control groups in terms of efficiency of regeneration. Wolff and Goodfellow[17] concluded that the course of hemoglobin concentration during the first 3 months of life was not improved by early administration of oral iron. Gaisford and Jennison[19] administered intramuscular iron (Imferon) to a group of premature infants during the third postnatal week and could demonstrate no difference between treated and control infants until the sixteenth week. Thus, there seems to be no convincing evidence the exogenously administered iron will accelerate the rate of hemoglobin gain if adequate iron stores are present.[12] As indicated above, the intermediate phase is variable in duration. Therefore, the question of whether an infant will or will not respond to iron depends not on the time that the medication is administered but rather on the state of the iron reserve at that time.

LATE PHASE

With the exhaustion of the iron stores laid down in the early phase and reutilized in the intermediate phase, the premature infant now becomes truly iron deficient. In the absence of additional iron from exogenous sources, a second fall in hemoglobin concentration occurs. Since iron deficiency primarily affects hemoglobin synthesis and only minimally involves red cell regeneration, the erythrocytes now develop morphologic and volumetric characteristics which permit the anemia in the late phase to be distinguished from that in the early and intermediate phases. That is, the anemia is now *hypochromic* and microcytic as compared with normochromic in the preceding phases.

As indicated before, the time of exhaustion of iron reserves will be dependent upon the variables involved in the intermediate and early phases. To repeat, iron depletion will be determined by the time at which iron reutilization begins which, in turn, will depend upon the time that stimulation for erythropoiesis occurs which, in turn, is regulated by the rate of fall in hemoglobin concentration in the early phase. Of all the factors involved, the initial circulating hemoglobin mass appears to be the most important single variable.

Fig. 4 demonstrates the manner in which a low initial hemoglobin mass may influence the time of depletion of iron reserves, the time of development of the late anemia, and, thus, the time when supplemental iron is required. It may be noted that the infant with a low initial hemoglobin mass has an early phase of only about 3 weeks in duration. Available iron reserves are exhausted by the age of 50 days and hemoglobin gain ceases. The second fall in hemoglobin is then seen. In contrast the infant with the higher initial hemoglobin mass does not begin reutilizing iron until 50 days of

age and has sufficient iron reserve to permit continuous hemoglobin synthesis for 120 days. In order to prevent the interruption of hemoglobin gain and the second fall in hemoglobin concentration, the first baby would have required supplemental iron at 6 to 7 weeks of age as compared with 16 to 17 weeks of age for the second baby. It should be noted that the weight curves are similar in these infants.

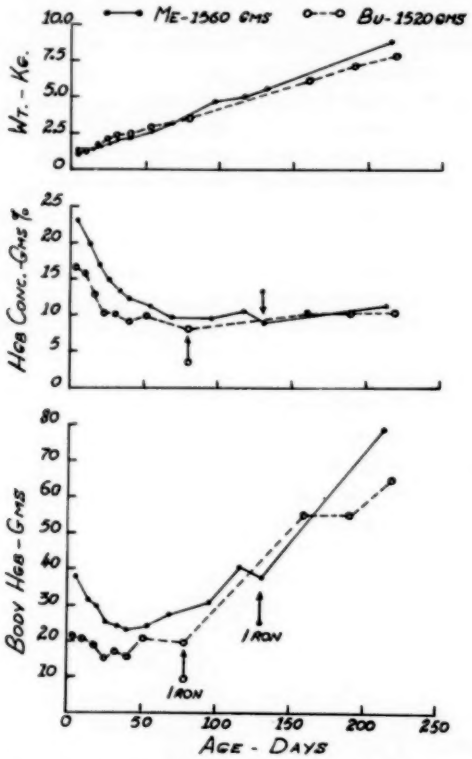


Fig. 4.—The role of initial hemoglobin level in determining the duration of the three phases of anemia and the time that supplementary iron is required.

It has become apparent that the iron needs for optimal hemoglobin synthesis are so great that an adequate supply of iron from dietary sources alone cannot be anticipated.[12] Thus, virtually all prematurely born infants require iron supplementation. Although the introduction of iron during the first month of life does not affect the early anemia of prematurity, it is well established that infants who have been receiving iron have significantly higher hemoglobin levels from the third to the fourth month onward than do untreated infants.[16-19] The exact time that supplementation is actually required is variable, as shown above, but very few babies will require iron before the sixth week, and virtually all will require iron by the fourteenth

to sixteenth weeks. From a practical standpoint, the institution of prophylactic iron administration at about 8 weeks of age should protect the great majority of infants. Estimates of iron requirements indicate that 15 mg. of elemental iron daily should be sufficient to prevent the late anemia of prematurity.[12] In instances where the late (iron-deficiency) anemia is already present, higher doses (50 to 100 mg. daily) are required. Because of the limited amounts of iron in food, untreated premature infants may manifest severe iron-deficiency anemia well into the second year of life, and most should continue receiving supplemental iron during the entire first year. The great tendency to iron deficiency in premature infants necessitates periodic checks of hemoglobin levels during the first 2 years.

Prevention of the late anemia of prematurity requires proper instruction of the parent and adequate facilities for follow-up and supervision. In areas where social and economic factors contribute to poor parental reliability and erratic follow-up, the incidence and severity of the late anemia in prematurely born infants is such as to constitute a public health problem of considerable magnitude. It is not at all uncommon for infants to be returned to the hospital during the second 6 months of life with hemoglobin levels below 4 Gm. per 100 ml. The physician caring for the premature infant is well aware of the population group served by his nursery and the community resources which are available. In addition the relatively long period of time that the infant remains in the nursery presents the opportunity for careful evaluation of the individual parent. In cases where the physician feels that the parent is not dependable and that adequate supervision will not be continued, the possibility of prophylaxis by means of parenteral iron administered prior to discharge should be considered. The availability of a safe, easily administered, completely utilized parenteral iron preparation makes it feasible to administer enough iron to prevent the late anemia. From 150 to 250 mg. of parenteral iron has been successful in this respect without apparent toxicity.[19] It should be emphasized that this is not recommended as a routine procedure but rather as a necessary preventive measure in situations where the occurrence of late iron deficiency is almost predictable.

DISCUSSION

During the early and intermediate phases of the anemia of prematurity, the infant exhibits a level of physiologic responsiveness which is extraordinary for so immature an organism. The regulatory hematopoietic mechanisms seem well established even in the smaller infants with birth weights less than 1,000 grams. Thus, the differences between the anemia of prematurity and the "physiologic anemia" of the full-term infant are essentially quantitative ones, and both appear to depend upon the same underlying pathogenetic events. The late anemias both in premature and in full-term infants, are based upon iron deficiency and demonstrate the rather precarious state of iron balance in all infants.[20] The very rapid rates of growth of the prematurely born infants and the relatively lower initial hemoglobin masses advance in time and exaggerate in degree the changes encountered in the full-term infant. With both groups,

the clinician is involved with a constantly changing organism. The proper management of these infants requires an intimate understanding of the nature of these changes.

CONCLUSIONS

The premature infant manifests anemia during three distinct phases. The early and intermediate phases depend upon variations in the rate of effective hematopoiesis and appear to be controlled by specific regulatory mechanisms. The anemias in these periods occur in the face of relative iron excess, are self limited, are essentially unaffected by prophylactic measures, and rarely require transfusions. The late anemia is based upon iron deficiency and may be effectively prevented by prophylactic administration of iron. However, the demands for optimal hemoglobin synthesis are such that an adequate supply of iron from dietary sources alone cannot be anticipated.

The phases of the anemia of prematurity occur in orderly sequence and are interdependent. Factors of birth weight, rate of gain, red cell longevity, and differences in hematopoietic responsiveness contribute to variations in timing and severity in the individual phases.

REFERENCES

1. Schulman, I., and Smith, C. H.: Studies on the Anemia of Prematurity. III. The Mechanism of the Anemia, A. M. A. J. Dis. Child **88:** 572, 1954.
2. Gairdner, D., Marks, J., and Roscoe, J. D.: Blood Formation in Infancy. IV. The Early Anaemia of Prematurity, Arch. Dis. Childhood **30:** 203, 1955.
3. Seip, M., and Halvorsen, S.: Erythrocyte Production and Iron Stores in Premature Infants During the First Months of Life. The Anemia of Prematurity—Etiology, Pathogenesis, Iron Requirement, Acta paediat. **45:** 600, 1956.
4. Metzroth, M.: Retikulocytenreifung und Lebensdauer der Erythrocyten bei Frühgeburten, Inaugural-Diss., Freiburg, 1956.
5. Mollison, P. L.: Blood Transfusion in Clinical Medicine, Springfield, 1956, Charles C Thomas.
6. Hollingsworth, J. W.: Lifespan of Fetal Erythrocytes, J. Lab. & Clin. Med. **45:** 469, 1955.
7. Gilardi, A., and Miescher, P.: Die Lebensdauer von autologen und homologen Erythrocyten bei Frühgebornen, reifen Neugebornen und Alterenkindern, Schweiz. med. Wchnschr. **87:** 1456, 1957.
8. Erslev, A. J.: Humoral Regulation of Red Cell Production, Blood **8:** 349, 1953.
9. Rooth, G., and Sjöstedt, S.: Hypoxia for Prevention of Physiologic Anemia in the Neonatal Period of Rats, Scandinav. J. Clin. & Lab. Invest. **9:** 174, 1957.
10. Seip, M.: Reticulocyte Studies; Liberation of Red Blood Corpuscles From Bone Marrow Into Peripheral Blood and Production of Erythrocytes Elucidated by Reticulocyte Investigations, Acta med. scandinav. (suppl. 282) **146:** 1, 1953.
11. Gairdner, D., Marks, J., and Roscoe, J. D.: Blood Formation in Infancy. II. Normal Erythropoiesis, Arch. Dis. Childhood **27:** 214, 1952.
12. Schulman, I.: Studies on the Anemia of Prematurity, *in* Iron in Clinical Medicine, Berkeley, 1958, Univ. of Calif. Press, p. 204.
13. Allen, E.: Unpublished Observations.
14. Schulman, I.: Characteristics of the Blood in Fetal Life, *in* Symposium on Oxygen Supply to the Human Fetus, Josiah Macy, Jr., Foundation, 1959, Oxford Univ. Press.
15. Langley, F. A.: Haemopoiesis and Siderosis in Foetus and Newborn, Arch. Dis. Childhood **26:** 64, 1951.
16. Reedy, M. E., Schwartz, S. O., and Plattner, E. B.: Anemia of the Premature Infant. A Two-Year Study of the Response to Iron Medication, J. PEDIAT. **41:** 25, 1952.
17. Wolff, J. A., and Goodfellow, A. M.: Hematopoiesis in Premature Infants With Special Consideration of the Effect of Iron and Animal Protein Factor, Pediatrics **16:** 753, 1955.
18. Sisson, R. T. C., and Whalen, L. E.: Meat in the Diet of Premature Infants. II. Influence on Red Cell Volume and Hemoglobin Mass, A. M. A. J. Dis. Child. **95:** 626, 1958.
19. Gaisford, W., and Jennison, R. F.: Intramuscular Iron in Infancy, Brit. M. J. **2:** 700, 1955.
20. Sturgeon, P.: Iron Metabolism. A Review With Special Consideration of Iron Requirements During Normal Infancy, Pediatrics **18:** 267, 1956.

HEINZ BODY ANEMIA AND RELATED PHENOMENA

CONRAD GASSER, M.D.
SWITZERLAND

ALTHOUGH Heinz bodies were described in 1890, until recent years they have been of little interest to the pediatrician since their appearance was usually associated with exposure to industrial toxins. The recent demonstration of hemolytic anemia with Heinz body formation in infants and children following exposure to a variety of poisons and drugs, including the administration of water-soluble vitamin K to premature infants, and the diagnostic significance of Heinz bodies in patients with agenesis of the spleen have made familiarity with this morphologic abnormality of more importance to the pediatrician.

Heinz bodies or Heinz-Ehrlich bodies are round, oval, or serrated granules and are situated close to the red cell surface. They can be easily demonstrated in unstained wet preparation by phase-contrast microscopy or by supravital stain with brilliant cresyl blue, nile-blue sulfate, methyl crystal violet, and many other dyes. These bodies cannot be demonstrated with Giemsa and Wright panoptic stains. When a supravital stain is used for a reticulocyte count, the Heinz bodies are also seen; when a fixed panoptic stain like May-Grünwald-Giesma is used as a counterstain, the reticuloeytes are restained but not the Heinz bodies. This method of counterstaining, first used by Zadek and Burg,[46] then by Dustin,[8] and also by us, allows the differentiation between the different kinds of granules in erythrocytes (Fig. 1).

From Table I it can be seen that chromatin remnants are stained by the Giemsa stain as well as by vital staining. The Heinz bodies as well as the marginal bodies of Röhl are only visible by vital staining. Heinz bodies must also be differentiated from siderocytes (Grüneberg) and the Pappenheimer bodies. Both of these red cell inclusion bodies give a positive iron staining with Berlin blue. The Heinz bodies are resistant to osmotic tests.

Heinz bodies do not appear in normal blood but may be seen in the blood of some normal premature infants during the first weeks of life. They, as well as the Howell-Jolly bodies, also occur with splenic agenesis or after splenectomy. Erythrocytes with Heinz bodies are damaged forms and should be segregated by the spleen. The persistent presence of Heinz bodies in the absence of the spleen is a rare condition and does not occur with anemia, cyanosis, or abnormal blood pigments, in other conditions.

TOXIC HEMOLYTIC HEINZ BODY ANEMIA

The presence of Heinz body formation in red cells is nearly always a sign of increased hemolysis and progressive anemia. Heinz body formation can be used as a screening test for poisoning by drugs and industrial chemicals. Since the discovery of

Heinz bodies over half a century ago, many substances have been known as Heinz body producers and the list continues to grow (Schilling,[34] Moeschlin,[26] Webster,[39] Buckell and Richardson,[4a] Gasser, Fertman and Fertman[10]).

TABLE I

	UNSTAINED WET PREPARATION (PHASE-CONTRAST MICROSCOPY)	SUPRAVITAL STAIN (BRILLIANT CRESYL BLUE; NILE-BLUE SULFATE; METHYL CRYSTAL VIOLET)	SUPRAVITAL COLORATION PLUS PANOPTIC STAIN (MAY-GRÜNWALD-GIEMSA)	PANOPTIC STAIN (WITHOUT PREVIOUS SUPRAVITAL COLORATION)
Nuclear remnants Howell-Jolly bodies Chromatin dust	+	+	+	+
Basophilic stippling Punctate basophilia	+	+	+	+
Reticulocytes Substantia reticulofilamentosa	–	+	+	–
Heinz bodies "Innenkörper"	+	+	–	–
Röhl bodies "Marginal bodies" Corpora marginalia	+	+	–	–
Iron bodies Siderocytes Grüneberg and Pappenheimer bodies	–	–	–	–

Of clinical importance is a small group of inorganic substances, namely, hydroxylamine, sodium chlorate, sodium nitrate, and sodium metasulfite. The main group consists of different kinds of aromatic substances:

1. aromatic amines, such as aniline and phenylhydrazine, as well as most sulfonamide drugs: aniline, acetanilid, paraminophenol (Phenacetin), phenylhydrazine, phenylsemicarbazide, acetylphenylhydrazine, sulfonamide, sulfapyridine, and sulfapyrimidine;

2. aromatic nitro derivatives: nitrobenzene (diaper-marking dye), dinitrobenzene, trinitrobenzene, trinitrotoluene, and paranitraniline ("parared" of wax crayon);

3. complex aromatic amino derivatives: nile-blue sulfate, methylene blue, and colchicine.

A special group is represented by the aliphatic and heterocyclic nitrogen containing compounds such as nitroglycerine, erythrol tetranitrate, and alloxantin. Of the small group of organic compounds without nitrogen, some are important for the pediatrician: guaiacol, resorcin, and napthalene (moth balls).

Heinz body anemia due to poisoning is occasionally encountered in the young infant, although the possibility of contact with industrial toxins is slight. Accidental poisoning occurs, for example, when special dyes are used to mark diapers, through medical application of skin lotions such as resorcin (see Fig. 2), or if the infant

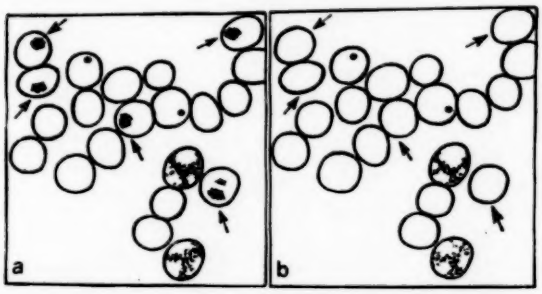


Fig. 1.—*Stain differentiation of reticulocytes, Howell-Jolly bodies and Heinz bodies. a, Brilliant cresyl blue stain:* Heinz bodies (small arrows), 2 Howell-Jolly bodies, and on the lower right 2 reticulocytes. *b, Same area, subsequently counterstained with May-Grünwald-Giemsa:* Heinz bodies invisible, reticulocytes and Howell-Jolly bodies can still be seen.

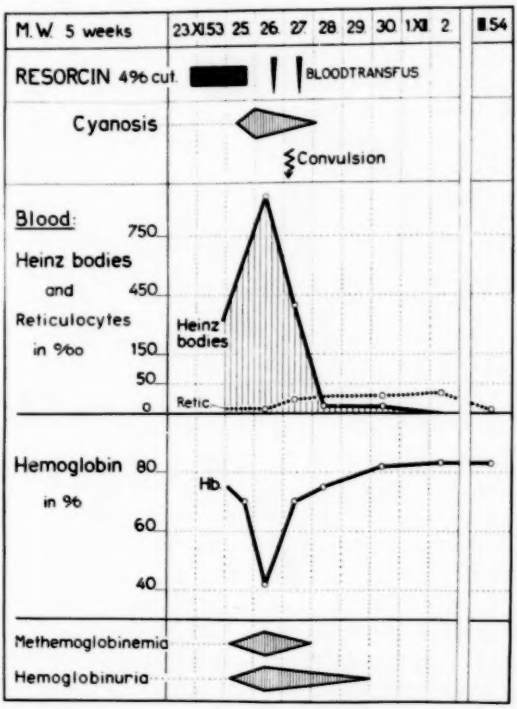


Fig. 2.—*Toxic hemolytic Heinz body anemia* in a 5-week-old infant with seborrheic dermatitis treated for 2 days with 4 per cent resorcin shake-lotion: 99 per cent Heinz bodies; severely damaged red cells (see Fig. 3 at right), hyperleukocytosis (4,5000), and monocytosis (6,850). Methemoglobinemia and hemoglobinuria. Clinically: cyanosis, pallor, slight jaundice, tachypnea, tachycardia, fever, vomiting, and convulsions. Increasing hepatosplenomegaly. Cured by two sizeable blood transfusions.

happens to eat moth balls containing naphthalene (Zuelzer and Apt[48]) or wax crayons containing paranitraniline. Heinz body anemia in infants was more often seen early in the sulfonamide era, because many of the early preparations were toxic and were strong Heinz body producers. The young infant is very susceptible to the toxic effects of sulfonamides. Certainly a great number of toxic Heinz body anemias have escaped diagnosis owing to the limited knowledge of this phenomenon.

Toxic Heinz body anemia is not limited to any age group or to a special disease, but there exists an individual predisposition to react with Heinz body formation. The degree of Heinz body formation does not correspond to the severity of the hemolytic anemia. We have seen high percentages with only slight hemolysis and, on the other hand, severe hemolytic anemia with slight Heinz body formation. There seems to be a relation between renal and hepatic funtion and the appearance of Heinz bodies, possibly owing to the important role of these organisms in neutralizing and eliminating toxins. Possibly hypoplasia of the spleen is also important.

Clinically, the most important early and obvious symptom is a gray cyanosis, the appearance of which should always be considered as a warning. The jaundice is usually not very significant, as the adequacy of hepatic function of young infants tends to avoid severe bile retention except in the newborn period. Quite often sudden pallor appears together with, or shortly after, cyanosis, depending upon the dose and the toxicity of the causative agent. The general condition deteriorates and the infant becomes rather drowsy and sometimes refuses feeding; fever may occur (Fig. 2).

Hematologic Data (Fig. 3).—As usual in acute hemolytic anemia, the Heinz body anemia is characterized by an increasing spherocytosis and a decreased osmotic resistance in vitro. The spherocytes have a tendency to burst, so that disintegrating erythrocytes show bizarre shapes which we call "broken egg shells." This typical morphologic picture can be seen in fresh unstained preparations as well as in fixed panoptic stained films. Similar morphologic pictures can be found in hemolytic-uremic syndromes and in favism. The key for the diagnosis is the proof of Heinz body formation in the red blood cells, as seen in the unstained fresh preparation by phase-contrast microscopy or more easily by pure supravital staining. The increased hemolysis leads to anemia of varying degree, bilirubinemia, and often methemoglobinemia and sulfhemoglobinemia (Fig. 2). The urobilin output in the stool and urine is increased. The signs of blood regeneration appear very soon after the beginning of the hemolysis, as evidenced by erythroblastosis of the bone marrow, a reticulocytosis in the peripheral blood, and sometimes a pseudoleukemic reaction with appearance of erythroblasts and myelocytes in the peripheral blood.

The differential diagnosis requires exclusion of other hemolytic anemias, which can be quite difficult if no supravital staining is carried out to detect the Heinz bodies. Sometimes leukemia has to be excluded because of a pseudoleukemic reaction and the

cyanosis may make one suspect congenital heart disease or pneumonia.

Treatment.—A blood transfusion is indicated to correct the anemia, and in severe cases an exchange transfusion may be considered to remove the causative agent. It is of greatest importance to detect the causative agent and to stop further contact with it.

The prognosis is nearly always favorable if the disease is recognized and treated promptly. If unrecognized, death can occur with continued administration of the toxic agent and/or through failure to provide blood by transfusion when it is needed.

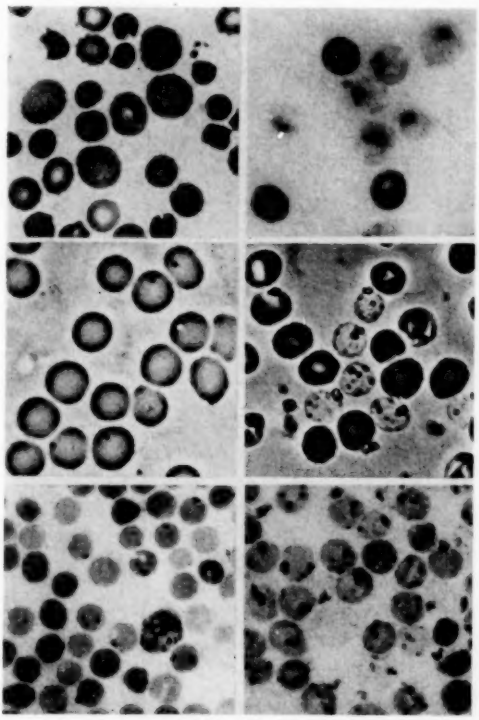


Fig. 3.—*Appearance of red cell in toxic Heinz body anemia. A, Panoptic stain (May-Grünwald-Giemsa):* At left, sulfa drug-induced acute hemolytic anemia with spherocytosis (in part, bursting cells), Heinz bodies not visible. At right, severe hemolysis due to resorcin resorption, most cells with washed-out hemoglobin, in the stromashadows darker spots, probably identical with Heinz bodies (unusual findings). *B, Phase-contrast microscopy (fresh unstained erythrocytes):* Heinz bodies visible as dark or bright spots (left) or occasionally as condensed irregular bodies at surface of stroma of the hemolyzed cells. *C, Supravital stain (brilliant cresyl blue):* Heinz bodies as single blue round bodies near the surface of the red cells (left), seldom as multiple spots (right). At left, below, a single reticulocyte.

SPONTANEOUS HEINZ BODY FORMATION AND ACUTE HEMOLYTIC HEINZ BODY ANEMIA IN PREMATURE INFANTS AND ITS RELATION TO VITAMIN K THERAPY

A slight degree of spontaneous Heinz body formation is not an abnormal finding in newborn infants, especially in premature ones. Willi[41] made

this observation initially in 1946 and subsequently (Willi and Hartmeier, 1950[44]) demonstrated that 18 per cent of 1,250 newborn infants had Heinz body formation in more than 20 per cent of their red blood cells; 84 per cent of this group were premature infants. This spontaneous Heinz body formation—also confirmed by us—need not be related to increased hemolysis or jaundice, and the phenomenon disappears within the first weeks of life.

In 1948 Gasser and Karrer[12] reported a case of fatal hemolytic anemia in a premature infant with the occurrence of Heinz bodies, marked destruction of the red blood cells, and severe jaundice. In 1951, two new cases were added, and in 1953 the author described the "hemolytic anemia of

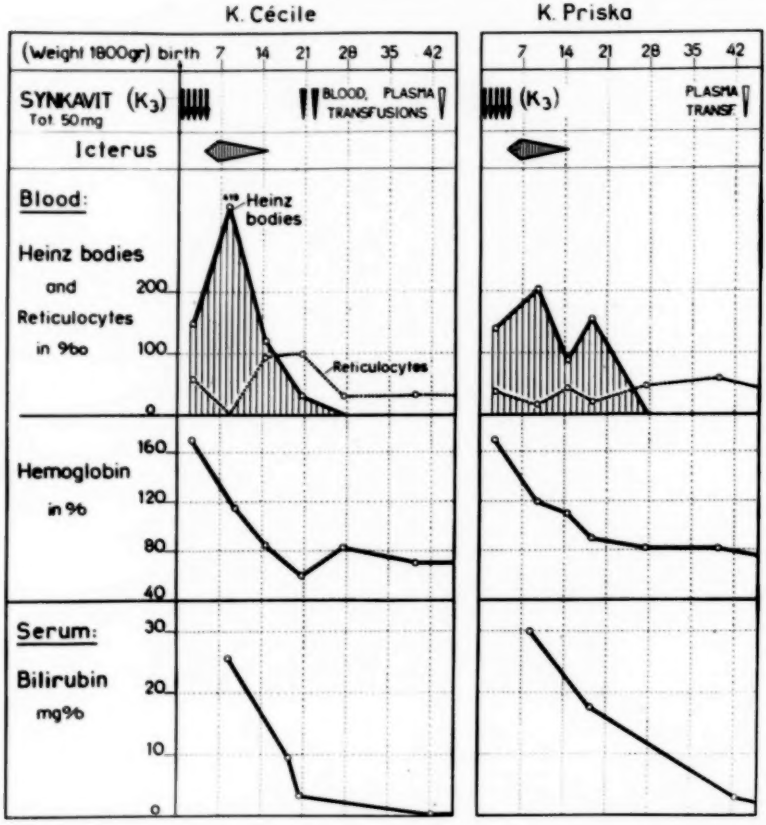


Fig. 4.—*Hemolytic Heinz body anemia of premature infants.* Observed in a pair of biovular twins, both weighing 1,800 grams, both received a daily injection of 10 mg. Synkavite (menadione diphosphate) during the first 5 days of life. Both showed simultaneous formation of Heinz bodies (maximum 419 per cent and 217 per cent, respectively) and severe jaundice (26 mg. per cent and 30 mg. per cent bilirubin, respectively). In the case plotted on the left, there was progressive anemia with poor general condition (lividness, drowsiness), and blood transfusions were required. The twin sister showed similar symptoms, but less severe. No transfusion was given, but the recovery was slower.

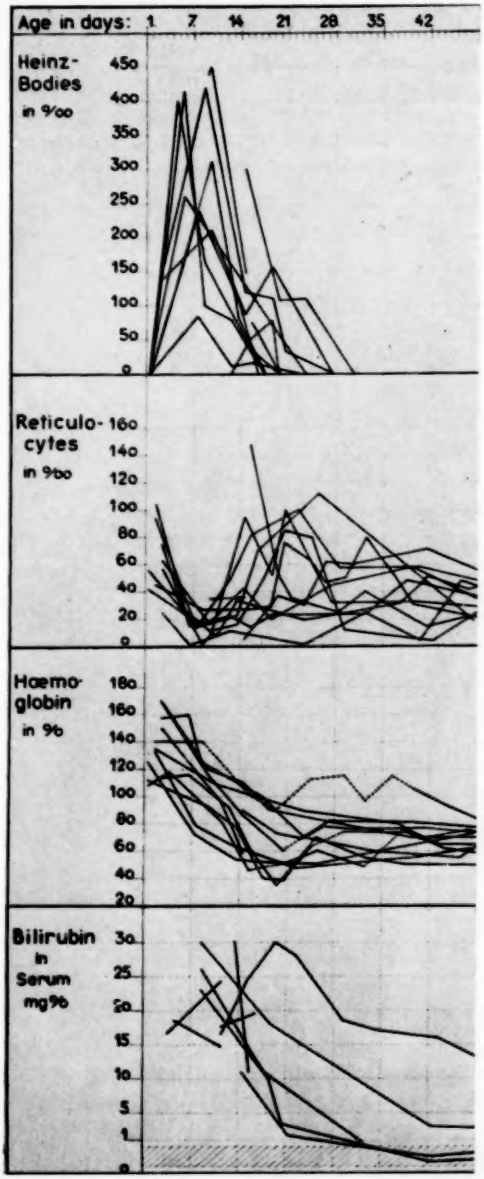


Fig. 5.—*Diagram summarizing the values of 14 premature infants with Heinz body anemia* which corresponds to the Heinz body peak appearing at the end of the first week. At this point one finds the lowest reticulocyte count and the highest bilirubin values. In the third week the anemia is pronounced, followed by reticulocytosis. (Dotted lines are hemoglobin values after blood transfusion.) These phenomena appear synchronized despite different doses and unequal duration of Synkavite medication.

premature infants with spontaneous Heinz body formation'' as a new syndrome, based on 14 personally observed cases with the following symptoms:

1. occurrence only in premature or underweight full-term infants;

2. severe jaundice, usually beginning after the first day, but often of long duration;

3. progressive anemia during the second to third week, usually coinciding with the decrease of the jaundice;

4. increased formation of Heinz bodies during the first to second week, preceding the anemia;

5. relative reticulocytopenia at the height of the formation of Heinz bodies, usually followed by reticulocytosis and occasionally erthroblastosis, without splenomegaly;

6. the clinical picture of generalized weakness, feeding problems, drowsiness, and failure to thrive;

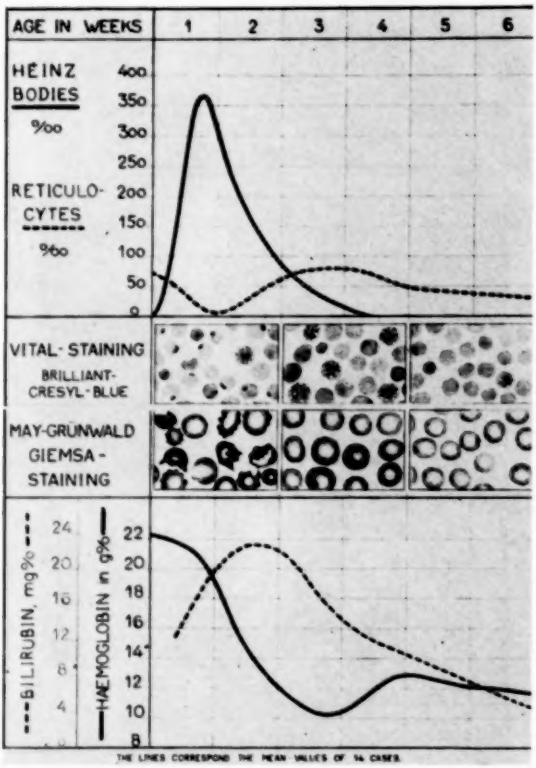


Fig. 6.—*Synthesis of Heinz hemolytic body anemia in premature infants.* The lines correspond to the mean values of 14 patients, as shown in Fig. 5, who were given Synkavite. The morphologic changes as seen in supravital stain (Heinz bodies and reticulocytosis) and in panoptic stain (disintegrating spherocytes) are also depicted.

7. absence of formation of specific immune antibodies in mother and child and absence of known toxic substances of exogenous origin, before, during, or after birth;

8. striking success of blood transfusion therapy.

All infants were ''premature'' by birth weights, which ranged from 1,480

to 2,220 grams. Some of them were twins born at term (Fig. 4). There were 8 boys and 6 girls. Malformations or asphyxia were not found. There was no anemia or jaundice at birth. Icterus appeared on the second or third day, increasing in the following days, with the peak at the end of the first or during the second week, and with a slow disappearance in the following weeks. As the jaundice disappeared, a progressive anemia appeared with pallor, general weakness, drowsiness, and refusal of food. The spleen was never strikingly palpable. The liver was slightly enlarged in some cases. There was no acholia of the stools and no edema. Only the first case terminated fatally.

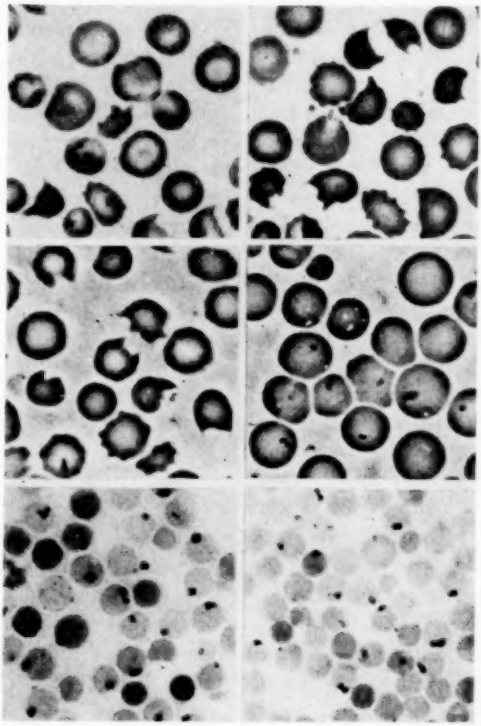


Fig. 7.—*Appearance of the red cell in Heinz body anemia of premature infants. A, Panoptic stain (May-Grünwald-Giemsa):* Two typical cases with disintegrating spherocytes of high hemoglobin content ("egg-shell" forms) next to the remaining normal macrocytes of the newborn infant. At left, chromatin remnants similar to Howell-Jolly bodies (no Heinz bodies). *B, Phase-contrast microscopy:* At left, irregularly shaped bursting erythrocytes, at right, Heinz bodies seen as dark and bright spots on the surface of the red cell. Both unstained wet preparations. *C, Supravital stain (brilliant cresyl blue):* In both cases irregularly shaped bluish-colored bodies in the erythrocytes.

Hematologic Data.—The principal finding was an increased Heinz body formation at the end of the first week of life and its disappearance in the second or third week, seldom later (see Figs. 5 and 6). At birth, no or only slight Heinz body formation was found, but in the first days high values were found. The mean value was 29.4

per cent. At the peak of Heinz body formation there was a reticulocytopenia and also an erythoblastopenia in the bone marrow, followed by a reticulocytosis and sometimes by slight erythroblastemia a few days later.

The morphologic changes of the red blood cells (Fig. 7) were characteristic and constant. As seen in fresh blood preparation by phase-contrast microscopy or in fixed preparation with panoptic stain, there was a pronounced tendency of the erythrocytes to burst after their metamorphosis to spherocytes, producing a marked polymorphia: normal macrocytes of the newborn infant mixed with broken irregularly shaped spherocytes, so-called "egg-shell" forms. In one case, chromatin remnants were also found (Fig. 7*A*). The Heinz body formation could be proved by phase-contrast microscopy as well as by supravital staining with brilliant cresyl blue. The Heinz bodies were similar to those found in toxic hemolytic anemia; they also disappeared by counterstaining the supravital preparation with May-Grüenwald-Giemsa stain. There was an unexpected sudden fall of the hemoglobin to an anemic level in the third week. The serologic findings in the mother and child showed no blood group incompatibility and no autoimmune-antibodies could be detected. The osmotic resistance was slightly decreased. The leukocyte count was mostly increased with neutrophilia and sometimes monocytosis. The platelets showed no abnormality in number or form. The bone marrow (examined in only a few cases) showed erythroblastosis after the onset of the anemia. In the blood serum, a considerable bilirubinemia (mean value of 21.4 mg. per cent) and a hypersideremia were found. There was no marked methemoglobinemia. The blood nonprotein nitrogen level was slightly increased in the first days of life.

Morphologic data are relatively rare because only a few cases were fatal. With our first patient who died at the height of the anemia, there was a marked hemosiderosis in the Kupffer cells of the liver, in the pulp cells of the spleen, and in the tubules of the kidney. Furthermore, an intensive phagocytosis of red blood cells was apparent in the liver and spleen. There was only slight extramedullary erythropoiesis and, accordingly, the weight of the liver and particularly of the spleen was low. The presence of giant cells in the adrenal cortex is worth mentioning.

THE PATHOGENESIS OF HEINZ BODY ANEMIA IN PREMATURE INFANTS

Between 1948 and 1953, we observed 14 premature infants with anemia and jaundice which could not be attributed to immune antibodies, to deficient regeneration (aplastic anemia), or to iron deficiency. The similarity between all of our cases was so striking (Fig. 5) that a common pathogenesis was considered. The syndrome occurred only in premature infants, was observed only during the first to third week of life, showed the hematologic findings described previously, and resulted in pronounced jaundice. In 1953 we discussed the pathogenesis and etiology of this syndrome and raised the question of whether an exogenous agent might not be involved, since the occurrence of massive Heinz body formation pointed to the possibility that toxic substances were acting upon the

blood of these premature infants. To study this possibility, the histories of the mothers were carefully reviewed to make sure that no common drug given before or during labor might have reached the infant through the placenta. The infants' charts were then reviewed. Only a water-soluble vitamin K derivative (Synkavite) in doses of 5 to 10 mg. daily had been given to all infants. Molitor and Robinson[27] (1940), Fromherz[11] (1941), Ansbacher, Corwin, and Thomas[2] (1942), and Cannavà[5] (1950) had previously commented upon the possible hemolytic effect of this hydroquinone derivative. Following our suggestion, Studer[36] (1953) showed that 5 mg. of Synkavite produced no Heinz body formation in mice, 10 mg. caused slight formation, and 50 mg. caused marked formation after 14 days. These experiments in mice showed that sublethal doses of water-soluble derivatives of menadione could lead to hemolysis and to Heinz body formation, whereas usual therapeutic doses seemed to be harmless and did not produce the phenomenon. Since then, we have omitted all prophylaxis

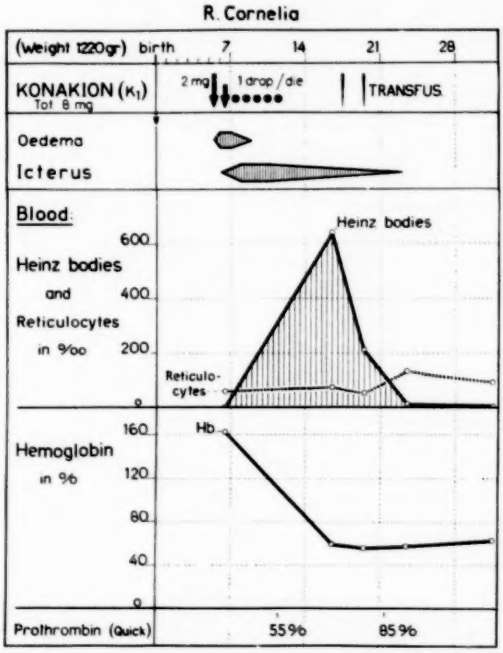
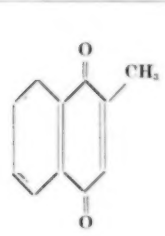


Fig. 8.—*Hemolytic Heinz body anemia in a premature infant* who received no Synkavite but received 8 mg. of *Konakion (vitamin K_1)*, distributed from the sixth to twelfth days of life. Birth, 2 months before term. Slight tendency to apnea, otherwise normal behavior. Received no other drug but Konakion and small doses of Lobeline (second to fifth days, total was 0.0024 Gm., given in 8 single doses of 0.0003 Gm. lobelinhydrochlorid). Edema and icterus gravis were followed by Heinz body formation and progressive anemia. The clinical and hematologic picture could be superimposed on the patients who received Synkavite. Successfully treated with blood transfusions. No kernicterus (courtesy of Dr. G. Schachenmann—common observation).

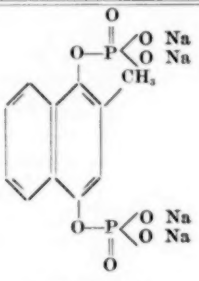
with Synkavite and other water-soluble vitamin K substitutes in newborn infants and have substituted vitamin K_1.

In 1955 Allison again drew attention to the possible danger of administering water-soluble vitamin K to premature infants after observing hemolytic Heinz body anemia and kernicterus. In 1956 Allison, Moore, and Sharman[1] showed that rats which had anemia in a small premature infant who received Synkavite. From 1956 through 1958, a number of authors discussed the probability that water-soluble vitamin K preparations produced hemolytic Heinz body anemia, severe icterus, and kernicterus (Meyer and Angus[25]; Zbinden, Schärer, and Studer[47]; Gleiss[17]; Sansone and Tomasseti-Levi[30]; Dyggve[8a]; Wespi[40]; Vest[38]).

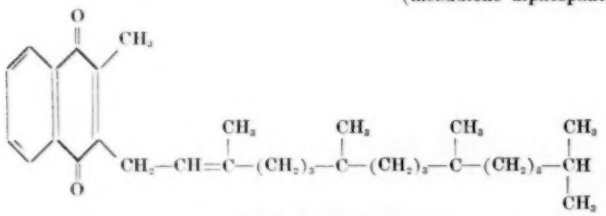
TABLE II



Menadione
(2 methyl-1,4 naphthochinon)



"Synkavite"
(tetrasodium 2-methyl-naphthalene-1,4 diol-diphosphate)
(menadione diphosphate)



Vitamin K_1 "Konakion"
2 methyl-3 phythyl-1,4 naphthochinon

been subjected to prolonged deficiency of vitamin E experienced severe hemolysis with hemoglobinuria and the occasional appearance of Heinz bodies when given Synkavite intramuscularly. Crosse, Meyer, and Gerrard[6] in 1955 also discussed the possibility that vitamin K substitutes might cause a higher incidence of kernicterus in premature infants. In 1956 Willi[43] observed a lethal case of Heinz body

The results of clinical investigations and experimental work with animals can be summarized in the following way: water-soluble derivatives of menadione, if given to premature infants in heretofore used doses, can produce a hemolytic anemia with Heinz body formation, hyperbilirubinemia, and kernicterus. The dose of such drugs for the premature infant should be reduced to 1 mg. daily, or the less

toxic vitamin K_1 substituted. Although vitamin K_1 is considered a harmless drug, we have observed a hemolytic Heinz body anemia in a premature infant who received 8 mg. of vitamin K_1 between the sixth and twelfth days of life. If others confirm this observation of Heinz body anemia in premature infants as a result of vitamin K_1 therapy, the whole problem of the administration of vitamin K to newborn infants, especially to premature infants, must be reconsidered.

It is not yet apparent why Heinz body anemia occurs after vitamin K therapy only in premature infants. In 1953 we discussed the possible roles of the spleen and liver. The red blood cell of the premature infant has a greater quantity of fetal hemoglobin but the type of hemoglobin seems to have no influence upon the occurrence of Heinz body anemia. Allison has discussed the possible role of vitamin E deficiency in the light of animal experiments by György and Rose[18] (1948-1950). The protective mechanism of the catalases is also significantly reduced in the newborn infant (Brenner and Allison[4]). However, the exact relationship of these many factors to Heinz body anemia is not clear.

HEINZ BODY PHENOMENON IN FAVISM

Recently, Sansone[31] (1957) proved the occurrence of Heinz body formation in a severe case of favism. The bodies could be seen in the unstained wet preparation by phase-contrast microscopy as well as by supravital staining with brilliant cresyl blue. They disappeared by counterstaining with May-Grünwald-Giemsa. The morphologic appearance of the red blood cells in fresh and fixed preparations was very similar to that of other types of Heinz body anemia. Heinz body formation seems to be a constant occurrence in the early state of the crisis in favism.

Sansone, Piga, and Segni[32] mention in their book on favism another important finding: a marked decrease and "instability" of the reduced glutathione of the blood (GSH) and a parallel and severe enzymatic deficiency of the erythrocyte involving glucose-6-phosphate dehydrogenase, even years after a crisis of favism had occurred. Favism may thus be compared to primaquine sensitivity, in which glutathione instability was also found (Beutler,[3] 1957). The relation between these findings and the Heinz body phenomenon may be of great importance for the pathogenesis of the latter. It is not the disease that is inherited, but rather the abnormal red blood cell on which the fava substances can act.

HEINZ BODY PHENOMENON IN AGENESIS OF THE SPLEEN

The triad of agenesis of the spleen, defects of the heart and large vessels, and situs inversus has been known to pathologists since the first description by Martin[24] in 1826. Gasser and Willi[14] independently made the clinical diagnosis of agenesis of the spleen on the basis of a spontaneous and continuous heavy formation of Heinz bodies in 1952 and in 1955[42] in 7 personally analyzed cases. In 5 of these the diagnosis was made during life. Four new cases were studied in the last years (Fig. 9), and in every case the constant formation of Heinz bodies led to the diagnosis which was later confirmed by autopsy. Our observations on the occurrence of Heinz bodies

in the absence of the spleen were based on Schilling's experiments[24] on animals. In 1912 he reported the appearance of Heinz bodies in splenectomized animals for the first time.

Clinical Findings.—Only two of the infants were premature and one of the others was underweight. No obvious malformations were observed at birth. The symptoms of the cardiac defect were cyanosis, tachypnea, and occasional collapse. Hemiparesis occurred in one patient. Cardiac signs were few, and in most instances the heart was not enlarged; the apex beat was normal (although reversed); the rhythm was regular; and the sounds were usually normal. A systolic murmur was heard only in a few patients. The electrocardiogram showed the typical tracings of situs inversus. In one patient there was also the typical pattern of the Wolff-Parkinson-White syndrome. Radiologically, those patients with cardiac involvement showed situs inversus with slight enlargement

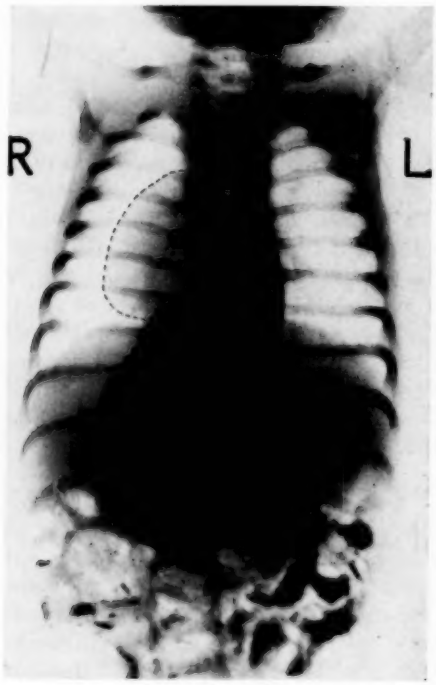


Fig. 9.

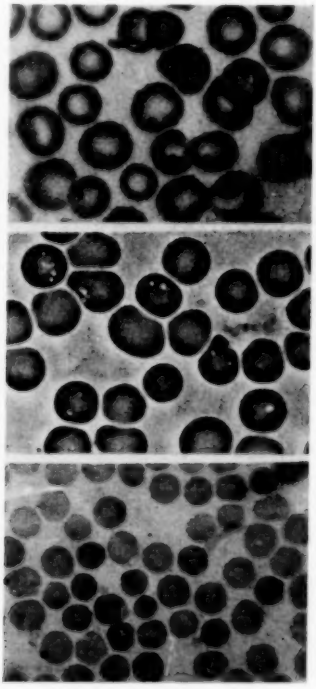


Fig. 10.

Fig. 9.—*Triad of spleen agenesis with its typical radiologic feature:* dextrocardia with the peculiar outline of the heart, showing a marked notch in the place of the pulmonary arch. Liver shadow demonstrates medial position. Stomach on the right, a sign of situs inversus abdominalis. Other abnormalities of the intestinal tract were found: relaxatio diaphragmatica and hernia paraesophageal diaphragmatica congenita. Absence of spleen. All findngs were confirmed by autopsy (unpublished case) (courtesy of Dr. Schachenmann—common observation).

Fig. 10.—*Hematologic findings* which make possible the intravitam diagnosis of spleen agenesis (same case as Fig. 9). *A, Panoptic stain (May-Grünwald-Giemsa):* Macrocytosis, erythroblastemia. *B, Phase-contrast microscopy* of wet unstained cells. Heinz bodies as intracorpuscular dark or bright granules, situated near the red cell surface. *C, Supravital stain (brilliant cresyl blue):* Heinz bodies as excentrically placed dark (bluish) spots.

of the heart (Fig. 9). The spleen could not be palpated, although an enlarged liver could be felt to both the right and left of the midline, owing to its median position. Barium contrast roentgenograms showed partial situs inversus abdominalis with gastric situs inversus being commonly present (Fig. 9).

Hematologic Data (Figs. 10 and 11).—Two hematologic changes are characteristic for the syndrome consisting of agenesis of the spleen, cardiac defects, and situs inversus. These are spontaneous and persistent presence of a high percentage of Heinz bodies in the erythrocytes and a variable but significant erythroblastosis in

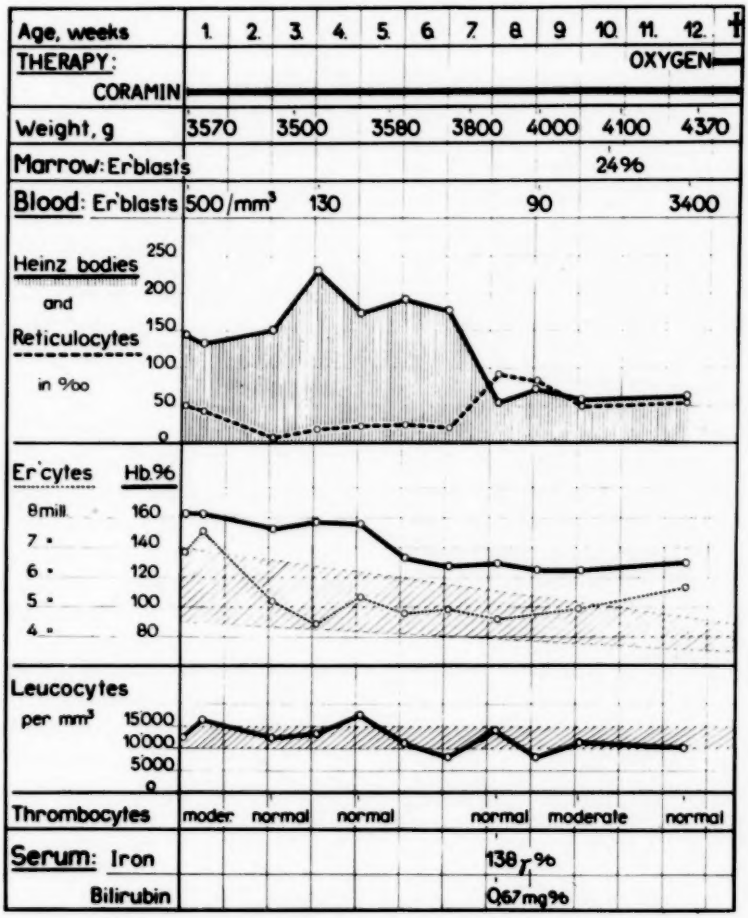


Fig. 11.—*Full-term newborn infant with normal body weight, displaying congenital cardiac disease with increasing cyanosis and dyspnea.* Death at the age of 3 months. On the basis of permanent high spontaneous formation of Heinz bodies, spleen agenesis was diagnosed. No anemia occurred and the morphologic picture of disintegrating red cells was not seen (case 4 of Willi and Gasser[42] 1955).

the peripheral blood. Furthermore, polyglobulia, macrocytosis, and the presence of chromatin residues in the erythrocytes are typical findings. The highest number of Heinz bodies was found during the first two months of life. The percentage of Heinz bodies varied between 11.2 and 20.0 per cent. They could be identified by the same methods as the toxic Heinz bodies, i.e., by phase-contrast microscopy in wet unstained preparation in which they appear as dark spots or brilliant vacuola-like spheres. In vital staining with brilliant cresyl blue, they could be seen as notched spherical inclusions on the cells' surface which disappeared after counterstaining with panoptic stain (May-Grünwald-Giemsa). The number of reticulocytes was slightly increased in all cases, but a true hemolytic crisis was never seen. The extent of erythroblastosis in the peripheral blood differed widely from infant to infant, and also fluctuated in each infant. The highest count was 40,000 per cubic millimeter, but the usual count varied between 100 to 1,200 per cubic millimeter. No proerythroblasts or megaloblasts were demonstrable. The erythrocyte counts were at the upper limit of normal or slightly higher. The hemoglobin values were proportionately higher, resulting in a high color index. As a rule, the erythrocytes were macrocytic and occasionally polychromatophilic with a slight tendency to become target cells. The Price-Jones curve was deviated to the right in all cases with an average diameter of from 7.5 to 8.7 μ. The hematocrit values were constantly high. The individual volume, the volume index, the hemoglobin contents, the thickness, the spheric index, and the saturation index were close to normal. The osmotic resistance tests showed an increased span with early initial hemolysis and increased terminal resistance. In all cases the occurrence of chromatin residuals in the form of Howell-Jolly bodies was noteworthy, but their number was far smaller than that of the Heinz bodies and never reached 5 per cent of the erythrocytes.

The leukocytes were moderately increased in number in some instances; in one case immature elements were present with a monocytosis. Thrombopoiesis was normal. In the bone marrow the erythroblasts were increased, especially in cases with marked peripheral erythroblastosis. There was no polymorphonuclear shift to the left, but marked karyorrhexis; no erythrophagocytosis was found. The serum iron level was slightly increased in most cases, as well as the bilirubin.

In 1955 we collected 46 cases of agenesis of the spleen. In 30 of these there was situs inversus associated with other malformations of the heart and/or great blood vessels. In the remaining cases there were malformations of the heart or situs inversus or neither of them. In four cases agenesis of the spleen was associated with various other defects. Agenesis of the spleen without other anatomic defects has a favorable prognosis and is rarely diagnosed, owing to lack of clinical symptoms. In individuals with cardiac malformations, the prognosis is always poor; the majority of them die during early infancy. Our oldest patient died at 3 years of age.

The cardiac malformations are nearly always identical: severe defects

of the septa which are either entirely absent or rudimentary. The atrioventricular valve leaflets are absent or rudimentary. Functionally, a biocular heart is observed in most instances. The origin of the large arteries is always modified; a pseudotruncus arteriosus, rotation anomalies of the heart, and large vessels and anomalies of the large veins are common. The situs inversus is only partial in most instances, involving the abdominal organs or often only individual organs.

The etiology is unknown. The malformations probably develop in the period in which the cardiac septum is formed and intestinal rotation occurs; that is, during the fourth to sixth weeks of fetal life.

SUMMARY

The erythrocytes of newborn infants and especially of prematurely born infants may contain a small number of Heinz bodies.

Heinz body formation and hemolytic anemia may occur following contact with a variety of drugs and poisons and following the administration of water-soluble vitamin K in large doses to premature infants.

The spontaneous appearance of Heinz bodies in older infants and children is associated with agenesis of the spleen.

My thanks are due to Prof. Fanconi, Kinderspital, Zurich, and to Dr. Gertrud Schachenmann, Pflegerinnenschule, Zurich, for their permission to publish data from their clinics as well as to Dr. Studer for his cooperation.

REFERENCES

1. Allison, A. C., Moore, T., and Sharman, I. M.: Hemolysis and Hemoglobinuria in Vitamin-E Deficient Rats After Injections of Vitamin-K Substitutes, Brit. J. Haemat. **2**: 197, 1956.
2. Ansbacher, S., Corwin, W. C., and Thomas, B. G. H.: Toxicity of Menadione, Menadiol and Esters, J. Pharmacol. & Exper. Therap. **75**: 111, 1942.
3. Beutler, E.: The Glutathione Instability of Drug-sensitive Cells, J. Lab. & Clin. Med. **49**: 84, 1957.
4. Brenner, S., and Allison, A. C.: Catalase Inhibition: A Possible Mechanism for the Production of Heinz Bodies in Erythrocytes, Experientia **9**: 381, 1953.

4a. Buckell, M., and Richardson, J. D.: The Significance of Heinz Bodies in the Erythrocyte, Brit. J. Indust. Med. **7**: 131, 1950.

5. Cannavà, A.: Azione metemoglobinizzante dei farmaci sintetici ad attività K vitaminica; ricerche sperimentali. Arch. "E. Maragliano" pat. e clin. **5**: 727, 1950.
6. Crosse, V. M., Meyer, T. C., and Gerrard, J. W.: Kernicterus and Prematurity, Arch. Dis. Childhood **30**: 501, 1955.
7. Debré, R., et al.: Anémies hémolytiques aiguës déterminées chez le petit enfant par l'administration de phényl-semicarbazide a doses élevées, Semaine hôp. Paris **26**: 1905, 1950.
8. Dustin, P., Jr.: Les corps de Heinz. Leur importance en hématologie et en pathologie expérimentale, Rev. belge sc. méd. **14**: 48, 1942.

8a. Dyggve, H.: Hematologic Studies in Premature Infants Who Received Menadione Derivates or Vitamin K, at Birth, International Congress on Hematology, Rome, 1958.

9. Fertman, M. H., and Doan, C. A.: Irreversible Toxic "Inclusion Body" Anemia, Blood **3**: 349, 1948.
10. Fertman, M. H., and Fertman, B. M.: Toxic Anemias and Heinz Bodies, Medicine **34**: 2, 1955.
11. Fromherz, K.: Pharmakologische Wirkungen von Vitamin K-Präparaten, Ztschr. f. Vitaminforsch. **11**: 65, 1941.
12. Gasser, C., and Karrer, J.: Spontan-"Innenkörperbildung" mit deletärer hämolytischer Anämie bei Frühgeburt, Schweiz. med. Wchnschr. **78**: 974, 1948.
13. Gasser, C.: Die hämolytischen Syndrome im kindesalter. Miteinem Geleitwort von Guido Fanconi, Stuttgart, 1951, Georg Thieme.
14. Gasser, C., and Willi, H.: Spontane Innenkörperbildung bei Milzagenesie, Helvet. paediat. acta **7**: 369, 1952.
15. Gasser, C.: Die hämolytischen Frühgeburtenanämie mit spontaner Innenkörperbildung; ein neues syndrom, beobachtet an 14 Fällen, Helvet, paediat. acta **8**: 491, 1953.
16. Gasser, C.: Perakute hämolytische Innenkörperanämie mit Methämoglobinämie nach Behandlung eines Säuglingsekzems mit Resorcin, Helvet. paediat. acta **9**: 285, 1954.

17. Gleiss, J.: Methyl-Napthochinon-Gaben und Gelbsucht bei unreifen Neugeborenen, Monatschr. Kinderh. **106**: 58, 1958.
18. György, P., and Rose, C. S.: Tocopherol and Hemolysis in Vivo and Vitro, Ann. New York Acad. Sc. **52**: p. 237.
19. Heinz, R.: Morphologische Veränderungen der roten Blutkörperchen durch Gifte, Virchows Arch. path. Anat. **122**: 112, 1890.
20. Heubner, W.: Methämoglobin, Innenkörper der Erythrocyten und Anämie, Klin. Wchnschr. **20**: 137, 1941.
21. Künzer, W., and Schütz, E.: Untersuchungen zur Heinzkörperbildung in Neugeborenenerythrocyten, Ztschr. Kindern. **77**: 101, 1955.
22. Laurence, B.: Danger of Vitamin K Analogues to Newborn, Lancet **1**: 819, 1955.
23. Levin, S. J.: Shoe-Dye Poisoning. Relation to Methemoglobin Formation, J. A. M. A. **89**: 2178, 1927.
24. Martin, G.: Bull. soc. anat. Paris, p. 39, 1841.
25. Meyer, T. C., and Angus, J.: The Effect of Large Doses of "Synkavit" in the Newborn, Arch. Dis. Childhood **31**: 212, 1956.
26. Moeschlin, S.: Versuche über die Entstehung von Innenkörpern in Erythrocyten (Heinzsche Körperchen) in vivo und vitro durch Sulfanilamid-Derivate und Phenylhydrazin, Folia haemat. **65**: 345, 1941.
27. Molitor, H., and Robinson, H. J.: Oral and Parenteral Toxicity of Vitamin K_1 Phthiocol and 2-methyl 1, 4, naphthoquinone, Proc. Soc. Exper. Biol. & Med. **43**: 125, 1940.
28. Moore, T., and Sharman, I. M.: Danger of Vitamin-K Analogues to Newborn, Lancet **1**: 819, 1955.
29. Polhemus, D. W., and Schafer, W. B.: Congenital Absence of Spleen; Syndrome With Atrioventricularis and Situs Inversus; Case Reports and Review of Literature, Pediatrics **9**: 696, 1952.
30. Sansone, G., and Tomassetti-Levi, A.: Sui rapporti fra vitamina K idrosolubile et anemia dell' immaturo a corpi inclusi, Minerva pediat. **9**: 39, 1957.
31. Sansone, G.: Patogenesi tossica del favismo ed alterata morfologia eritrocitaria, Minerva med. **40**: 3317, 1957.
32. Sansone, G., Piga, A. M., and Segni, G.: Il Favismo, Torino, 1958, Edizioni Minerva Medica.
33. Schachenmann, G.: Personal Communication.
34. Schilling, V.: Erweiterte hämatologische Verwertung des dicken Bluttropfens für Kernkugeln, Innenkörper, Endothelien, etc., Deutsche med. Wchnschr. **47**: 825, 1921.
35. Schulman, I., Smith, C. H., and Stern, G. S.: Studies on the Anemia of Prematurity, A. M. A. Am. J. Dis. Child. **88**: 567, 1954.
36. Studer, A.: Personal Communication (1953).
37. Varadi, S., and Hurworth, E.: Heinz Body Anemia in the Newborn, Brit. M. J., p. 315, 1957.
38. Vest, M.: Der Einfluss von Naphthohydrochinonderivaten (wasserlöslichen Vitamin-K-Ersatzpräparaten, Synkavit) auf Erythrocytenabbau und -regeneration bei Frühgeburten und auf das Glukuronidbildungsvermögen der Leber in vitro, Schweiz. med. Wchnschr., p. 969, 1958.
39. Webster, S. H.: Heinz Body Phenomenon in Erythrocytes, A Review, Blood **4**: 479, 1949.
40. Wespi, H. J.: Synkavit-Ikterus und Vitamin-K-Prophylaxe beim Neugeborenen, Gynaecologia **145**: 341, 1958.
41. Willi, H.: Innenkörperbildung durch Elkosin und spontane Innenkörperbildung, Schweiz. med. Wchnschr. **77**: 243, 1947.
42. Willi, H., and Gasser, C.: The Clinical Diagnosis of the Triad Spleen Agenesis, Defects of the Heart and Vessels and Situs Inversus, Neo-Natal Studies **4**: 25, 1955.
43. Willi, H.: Synkavit-Schädigung bei Frühgeborenen, Helvet. paediat. acta **11**: 325, 1956.
44. Willi, H., and Hartmeier, F.: Spontane Innenkörperbildung beim Neugeborenen, Schweiz. med. Wchnschr. **80**: 1091, 1950.
45. Willi, H., Vest, M., Käser, O., and Scheier, F.: Das hämatologische Verhalten der Frühgeborenen unter der Einwirkung hoher und niederer Dosen von Vitamin K_1 (Konakion), International Kongress Hämatologie, Rome, 1958.
46. Zadek, I., and Burg, K.: Innenkörperanämien, Folia haemat. **41**: 333, 1930.
47. Zbinden, G., Schärer, K., and Studer, A.: Experimentelle Untersuchungen über Erythrocytenschädigung durch Menadionderivate im Vergleich zu Vitamin K_1, Schweiz. med. Wchnschr., p. 1238, 1957.
48. Zuelzer, W. W., and Apt, L.: Acute Hemolytic Anemia Due to Naphthalene Poisoning, J. A. M. A. **141**: 185, 1949.

LEUKEMIA IN THE NEWBORN INFANT

MILA I. PIERCE, M.D.
CHICAGO, ILL.

LEUKEMIA in the newborn infant presents several distinctive features of etiological, clinical, and diagnostic interest. Although the pathogenesis of congenital leukemia is as obscure as it is in the childhood form, the existence of the disease in utero suggests the role of maternal and genetic factors which may prove to be etiologically significant in both forms. The predominance of the myelogenous type of leukemia in the newborn infant has contributed to the difficulty in establishing a clinical diagnosis, since the findings are simulated so closely by a leukemoid reaction to infection and by the hemolytic and hypoplastic anemias of the newborn infant. The pathognomonic criteria of the disease, namely: (1) the proliferation of certain precursor cells of the blood leukocytes and (2) the infiltration of these immature cells into extrahematopoietic tissues, must be demonstrated in the newborn infant before the diagnosis of congenital leukemia is acceptable.

CASE REPORTS IN THE LITERATURE

Leukemia is a rare disease in the newborn infant; the total number of cases recorded in the literature in sufficient detail to establish a diagnosis is limited to 45 cases (see Table I). In the first review of congenital leukemia in 1939, Kelsey and Anderson[1] accepted 8 cases (cases 1 to 8 in Table I) from the literature on the basis of an onset of symptoms within the first month of life, the clinical findings of hepato- and splenomegaly, a hyperleukocytosis with a preponderance of immature cells in the peripheral blood, and the histopathological findings of extensive infiltrations which could not plausibly have occurred in extrauterine life. They rejected 8 previously reported cases for lack of sufficient data and added one report (case 9) of their own. In 1951 Bernhard and co-workers[10] collected 6 more reports from this country (cases 10 to 15) and added 4 cases of their own (21 to 24). More than one-half of the total number of cases have been recorded within the past 10 years. Houstek and Brachfeldova[16] reported 3 cases (16 to 18). Also several single reports appeared in this country (cases 19, 20, and 25 to 28) and abroad (32 to 36) between 1950 and 1956. Hjelt and Wegelius[23] reported 4 cases from Finland, 3 of which (29 to 31) presented signs of the disease in the neonatal period. Shindo[25] reported 6 cases from Japan, of which 3 (33 to 35) were diagnosed in the newborn period. In a nationwide questionnaire survey designed to determine the frequency of the simultaneous occurrence of leukemia and mongolism, Krivit and Good[27] collected 10 cases of leukemia in mongoloid infants (cases 25 and 37 to 45).

TABLE I. CASES OF CONGENITAL LEUKEMIA REPORTED IN THE LITERATURE

CASE	AUTHOR AND REFERENCE	YEAR REPORTED	PREVIOUSLY REVIEWED
1	Smith[2]	1921	
2	Koch[3]	1922	
3	Stransky[4]	1925	
4	Bungeler[5]	1931	Kelsey and Anderson[1]
5	White and Burns[6]	1931	
6	Giblin[7]	1933	
7	Kornmann[8]	1934	
8	Cooke[9]	1939	
9	Kelsey[1]	1939	—
10	Morrison et al.[11]	1939	
11	Poncher[12]	1942	
12	Cross[13]	1944	Bernhard et al.[10]
13	Cross[13]	1944	
14	Hamme[14]	1944	
15	Keith[15]	1945	
16	Houstek and Brachfeldova[16]	1949	—
17	Houstek and Brachfeldova[16]	1949	—
18	Houstek and Brachfeldova[16]	1949	—
19	Heen[17]	1950	—
20	Taylor and Geppert[18]	1950	—
21	Bernhard et al.[10]	1951	—
22	Bernhard et al.[10]	1951	—
23	Bernhard et al.[10]	1951	—
24	Bernhard et al.[10]	1951	—
25	Casilli et al.[19]	1952	—
26	Potter[20]	1952*	—
27	O'Connor et al.[21]	1954	—
28	Reimann et al.[22]	1955	—
29	Hjelt and Wegelius[23]	1956	—
30	Hjelt and Wegelius[23]	1956	—
31	Hjelt and Wegelius[23]	1956	—
32	Fischer[24]	1956	—
33	Shindo[25]	1956	—
34	Shindo[25]	1956	—
35	Shindo[25]	1956	—
36	Strenger[26]	1956	—
37	Farber[27]	1956	
38	Krivit and Good[27]	1956	
39	Krivit and Good[27]	1956	
40	Schunk and Lehman[28]	1954	
41	Smith	—	Krivit and Good[27]
42	Wolf	—	
43	Andrea	—	
44	Des Forges	—	
45	Lee[29]	1957	

*Case 26 was cited by author.

These cases are all reported as congenital leukemia, and they can be separated into three groups: (1) infants exhibiting clinical signs of the disease *at birth*, (2) infants who appeared normal at birth but who developed clinical and hematologic signs within the newborn period, and (3) infants in whom the diagnosis was not established until the third to the sixth week of life.

The cases listed in Table II were infants who presented signs of leukemia at birth. The majority of these infants exhibited signs of severe systemic involvement and died of the disease within a few days. On delivery many of the infants appeared pale or livid and showed a variable degree of respiratory distress due to pulmonary infiltration or atelectasis. The liver and spleen were enlarged, although in 3 cases splenomegaly was absent at birth. Lymphadenopathy was not a constant finding. Hemorrhagic manifestations occurred in all, ranging from scattered petechiae to confluent ecchymoses in the skin and bleeding from the umbilical stump or from the mucous membranes of the gastrointestinal or genitourinary tracts. The absence of jaundice is noteworthy in view of the constant occurrence of hepatomegaly. Nodular skin infiltrates were described in 10 of the 21 infants exhibiting other signs of leukemia at birth and in only 1 of the infants in which the disease became manifest after birth. Two of the cases (2 and 3) with skin nodules were stillborn premature infants. The characteristic lesion was described well by Fortina and Petrocini[30] as a slightly raised nodule, a bluish to slate-gray color, which is palpated as a woody lump in the deep layers of the skin, and which is freely movable over the subcutaneous tissue. The overlying epidermis is atrophic and may ulcerate later. The nodules are multiple and scattered widely over the surface of the body.

The essential hematologic findings at birth were the hyperleukocytosis and predominance of immature granulocytes, characteristic of leukemia in the peripheral blood. It is significant that all the cases diagnosed at birth were reported as granulocytic or myeloid leukemias. The percentage of myeloblasts in the peripheral blood varied in the different infants from 10 to 80 per cent, but the majority of the infants showed a predominance of promyelocytes and myelocytes, identifying the leukemia as granulocytic rather than lymphoid. The number of platelets was reduced and the bleeding time was prolonged in the several infants in whom these values were reported. It is noteworthy that anemia was not a prominent symptom at birth, although the pallor and respiratory difficulty were common. In all of the infants in whom the hemoglobin and erythrocyte levels were determined shortly after birth, the values were within limits of normal but fell rapidly thereafter. The absence of anemia at birth is in sharp contrast to the regular occurrence of anemia in cases which are diagnosed in the later days of the newborn period.

The duration of life in the liveborn infants presenting signs of leukemia at birth was less than 2 weeks in 8 of 21 cases and less than 2 months in the remainder of the group. The course of the disease was marked by

TABLE II. CONGENITAL LEUKEMIA CASES WHICH EXHIBITED

CASE	AUTHOR AND REFERENCES	YEAR REPORTED	MATERNAL HISTORY					CLINICAL COMMENT	SEX	AGE FIRST SYMPTOM WAS NOTICED
			AGE	PARA	UTERINE BLEEDING	PELVIMETRY	SEROLOGY			
1	Smith[2]	1921	?	IV	?	?	Neg.	Enlarged axillary nodes at 5 weeks; hepatosplenomegaly at 6 weeks	M	3 wk.
2	Koch[3]	1921					Neg.	Stillborn premature; marked infiltration of skin, breast, genitalia, and organs		
3	Stransky[4]	1925						Skin infiltrates at birth	M	Birth
4	Bungeler[5]	1931		III			Neg.	Stillborn premature 7 months	M	
10	Morrison et al.[11]	1939					Neg.	Skin infiltrates at birth	F	Birth
14	Hamme[14]	1944				Neg.	Neg.	Purpura, severe at birth; proptosis; family history negative	F	Birth
16	Houstek and Brachfeldova[16]	1949	?	II	?	?	Neg.	Purpura at birth	M	Birth
17	Houstek and Brachfeldova[16]	1949					Neg.	Skin nodules at birth		Birth
20	Taylor and Geppert[18]	1950	?	II	0*	0	Neg.	Skin nodules at birth	M	1 day
21	Bernhard et al.[10]	1951	37	III	?	?	Neg.	Mongol	M	Birth
22	Bernhard et al.[10]		25	?	?	?	Neg.	Klippel-Feil syndrome	F	Birth
23	Bernhard et al.[10]		?	?	?	?	Neg.	Skin nodules at birth	M	Birth
24	Bernhard et al.[10]		?	?	?	?	Neg.	Radii absent	M	Birth
26	Potter[20]	1952	39	V	0	0	Neg.	Skin nodule at birth	F	Birth
28	Reimann et al.[22]	1955	?	II	?	?	Neg.	Skin nodule at birth		Birth
29	Hjelt and Wegelius[23]	1956					Neg.	Twin; spleen not enlarged at birth	F	Birth
30	Hjelt and Wegelius[23]	1956					Neg.		M	Birth
32	Fischer[24]	1956	?	I			Neg.	Skin nodules at birth	M	Birth
35	Shindo[25]	1956					Neg.	Petechiae at birth; diagnosed on forty-ninth day	F	Birth
	Strenger[26]	1956	—	—	—	—	Neg.	Petechiae and skin nodules at birth	F	Birth
40	Schunk and Lehman[28]	1954	38	III			Neg.	Mongolism; Rh incompatible		Birth

*+ = slight; ++ = moderate; +++ = severe; ++++ = very severe; 0 = not present.

SIGNS OF DISEASE AT BIRTH: SUMMARY OF CASE REPORTS

AGE AT DEATH	TYPE OF LEUKEMIA	JAUNDICE	PURPURA	MELENA	HEPATO- AND SPLENOMEGALY	SKIN INFILTRATION	HGB. (GM. %)	RBC	WBC	PLATELETS	BLASTS (%)	POST-MORTEM
2 mo.	Lymphatic	?	+*	0	++ +++	0	—	—	189,000	—	97	None
Stillborn	Myeloid	0	+	0	++ ++	+	—	—	—	—	—	Yes
3 wk.	Myeloid	0	+	0	++ ++	+	85	4.00	181,000	—	86	Yes
Stillborn	Myeloid	—	+	0	+++ +++	+	—	—	—		80	Yes
10 da.	Myeloblastic	0	+++	Via vagina	+++ +++	+	100	300,000			67 Myeloblasts in bone marrow	
20 da.	Myeloid		+	+	+ +							Infiltration into spinal cord
4 da.	Myeloblastic	+	+++	0	+++ +++	+++	117	4.20	82,400	60,000	73.9	Yes
26 da.	Micromyeloblastic	0	++	0	+++ +++	++	110	6.00	135,000	—	88	No
4 da.	Promyelocytic	0	+	Hematemesis No lymph node enlargement	+++ +++	++++	94	4.18	190,000	Red	60	Yes
1 da.	Myeloid	0	0		++ ++	0	15.3	4.20	246,000		25	Yes
3 wk.	Myeloid	0	+	Hema-	++ ++	0	104	5.90	32,000		30	Yes
7 da.	Myeloid	0	+	temesis	++ ++	+	—	4.20	198,000		15	Yes
2 mo.	Myeloid	0	+		++ ++	0	—		12,000		—	Yes
56 hr.	Myeloid	0	++	0	+++ +++	++	14.5	14.5	369,000	Red	89	Yes
14 wk.	Myeloid	0	0	0	0 0	0	111	4.9	6,900	normal ?	?	
				Later	+++ +++			3.2	89,000	Red		Yes
20 da.	Myeloblastic	0	+	0	+ +	−	3.6		2,800	Red	—	Yes
5 mo.	Myeloblastic	0	+	0	+ +	−	6.5		17,000	Red	—	Yes
1 da.	Myeloblastic	0	+	0	+ +	−	94	4.6	5,900	300,000	44	Yes
59 da.	Lymphatic	0	+	0	+ +	−	34	1.75	200,000	12,000	97	Yes
22 da.	Paramyeloblastic	0	+	0	+++ +++	+	98	3.60	199,000	12,000	49	Yes
11 da.	Myeloid	0	++	0	++ ++	0	12.5		100,000			Yes

progressive signs of organ infiltration, hemorrhage into the gastrointestinal tract or lungs, and death was commonly due to atelectasis or pulmonary hemorrhages.

The following case history is an example of this congenital type. It was the only case of leukemia in a newborn infant among 36,529 babies delivered at Chicago Lying-in Hospital between July 1948, and 1958, and was recorded by Potter[20] in 1952.

CASE REPORT

The girl was a product of the fifth pregnancy of a 39-year-old mother whose general health had been excellent throughout the pregnancy. An appendectomy was done on the mother when she was 16 years old, and at the age of 28 and again at the age of 29 years fibrous tumors were removed from her right breast. All four of her previous pregnancies were uncomplicated, and she had never been subjected to pelvimetry. All of the previous siblings were normal at birth. Three children were living and well; the second child died at four years of age of diphtheria. Throughout the current pregnancy she was attended in the prenatal clinic of the Chicago Lying-in Hospital; no severe upper respiratory infections or vaginal bleeding had occurred.

The infant was born after 4½ hours of normal labor which terminated naturally. At birth, the infant appeared to be healthy and cried spontaneously. The birth weight was 2,765 grams, the length was 49 cm. There were no signs of external bleeding; however, multiple subcutaneous purple nodules were scattered over the entire body including the extremities and scalp. The largest of these lesions measured 3 by 2 by 1 cm., the majority ranging between 0.5 and 1.0 cm. in diameter. Most of the lesions were flat and firm, the larger ones were slightly raised. The skin was freely movable over the underlying tissues, and the nodules were not adherent to underlying structures or bones. A few scattered petechiae were present on the trunk and extremities, and ecchymoses were present at the sites of pressure on the head. No deformities of the skull or bones were detected. The pupils were reactive and the extraocular movements were normal. Ecchymoses were present in the bulbar conjunctivae of the left eye. The optic fundi were well visualized, no retinal hemorrhages were detected. The buccal mucous membranes were intact; hemorrhagic and petechial signs were noted on the buccal surface of the upper lip. The examination of the eyes and nose revealed no abnormalities. The lungs were incompletely expanded, diminished resonance was detected over both upper lobes. The heart rate was 140 per minute, no murmurs were detected, the rhythm was regular. The liver was enlarged, the lower margin reaching 3 cm. below the costal margin in the right anterior axillary line. The spleen tip was palpable 1 cm. below the costal margin at the left anterior axillary line. The genitalia were normal; no bleeding from the vagina was present. No skeletal abnormalities were detected.

The hematologic findings revealed: hemoglobin 14.5 Gm. per cent; erythrocytes 4.75 million; leukocytes 369,000; the platelets were reduced and obscured by the large number of leukocytes on the smear; reticulocytes 0.45 per cent; hematocrit 45 per cent; bleeding time 3 minutes. The differential count of 300 leukocytes was: eosinophils 0.5 per cent, promyelocytes and early myelocytes 89.7 per cent, late myelocytes 0.3 per cent, segmented neutrophils 0.7 per cent, lymphocytes 1.0 per cent, large blasts 3 per cent, histiocytes 2 per cent, basophilic erythroblasts 0.2 per cent, normoblasts 2 per cent. Oxidase-positive cells were present in a ratio of 2:1 as compared to the oxidase-negative cells.

The general condition of the infant continued to be fairly good until the second day of life when respirations became labored and attacks of cyanosis developed. The size of the liver and spleen increased, but the level of the hemoglobin remained at 14 Gm. per cent. At 56 hours of age the infant died suddenly in an attack of respiratory distress.

The postmortem examination was conducted by Dr. Edith Potter who made the following comments: "Infiltrations composed of cells similar to those in the peripheral blood were found in the skin, connective tissue, muscles, and organs. The lymphoid tissue was reduced; the thymus weighed 4.7 grams, the spleen 13.2 grams. The malpighian corpuscles of the spleen were obliterated by the hyperplasia of immature leukocytes in the pulp and sinuses. The liver was enlarged and weighed 220 grams. A wide band of immature cells were present in the portal triads. The liver substance was heavily infiltrated throughout. Infiltrates were not found in the thyroid, adrenals, or brain substance. The dermis was widely infiltrated, and in the areas of gross nodules the infiltration by immature cells obliterated the normal local structures."

In the group of patients (Table III) who developed signs of disease within the first few days or within three weeks of life but in whom no detectable signs of leukemia were noted at birth, the symptoms of illness were ill defined and inconstant or absent. Failure to gain, diarrhea, low-grade fever, and skin rashes were noted. The occurrence of petechiae was the heralding sign of the disease in 9 cases. Progressive hemorrhagic signs developed in all cases and several developed melena as an early sign. Splenomegaly was not a constant finding but a variable degree of hepatomegaly was noted in all cases. Lymphadenopathy was rarely noted.

The hematologic findings were not recorded regularly at the detection of the first signs of illness, but anemia was present in all patients when the first blood count was taken after the fifth day of life. The anemia was normochromic and nonhemolytic. The degree of leukocytosis varied considerably; case 18 showed a leukocytosis of 855,000 on the third day of life; in case 19 the white blood cell count was only 23,000 on the fifth day. In all cases which were followed by serial leukocyte counts the levels rose progressively before death. Again the myeloid type of leukemia predominated in this group of infants as it did in the group which showed signs of the disease at birth. The percentage of myelocytes and myeloblasts varied from 15 to 90 per cent, and monocytoid blasts were noted in several cases reported as myelocytic leukemia. Micromyeloblastic leukemia was reported once (case 18) and leukemic reticulosis once (case 39). Three cases (5, 19, and 30) were reported as the lymphatic type. Only one (case 45) was reported as the stem-cell type. Case 27, reported by O'Connor and co-workers,[21] is one of the few in which the diagnosis of leukemia could be accepted in the presence of a septicemia. This newborn infant was a mongoloid who was delivered in the obstetrical unit of the hospital. At birth he showed signs of cyanosis, congenital heart disease, and hepatomegaly, but no hemorrhagic signs. The cyanosis improved, and he was allowed to go home with his mother on the third day. However, he was readmitted to the pediatric unit at 5 days of age with signs of extreme illness, cyanosis, and deep jaundice, and a hyperleukocytosis with a predominance of 90 per cent blasts of the myeloblastic type, a thrombocytopenia, but no anemia. He died within 2 hours after admission. A positive blood culture for hemolytic streptococci was obtained from the cultures of heart blood at autopsy. Leukemic infiltration of blast-like cells was extensive in the myocardium, pericardium,

TABLE III. CASES OF LEUKEMIA WITH ONSET OF SYMPTOMS

CASE	AUTHOR AND REFERENCES	YEAR REPORTED	MATERNAL HISTORY					CLINICAL NOTE	SEX	AGE FIRST SYMPTOM WAS NOTED
			AGE (YEARS)	PARA	UTERINE BLEEDING	RADIATION	SEROLOGY			
5	White and Burns[6]	1931					Neg.	Purpura on fourth day; hepato- and splenomegaly at 3 weeks	M	4 da.
6	Giblin[7]	1933					Neg.	Splenomegaly at 2 weeks	M	2 wk.
8	Cooke[9]	1939						Lump appeared on sternum at age 8 days		10 da.
18	Houstek and Brachfeldova[16]	1949		I			Neg.	At 15 days of age diarrhea and hemorrhagic signs		3 wk.
19	Heen[17]	1950		I			Neg.	Vaginal bleeding on fifth day; purpura on fourth day; staphylococcus sepsis	F	5 da.
25	Casilli et al.[19]	1952		I			Neg.	Skin lesions on eighth day; first examination twelfth day	F	8 da.
27	O'Connor et al.[21]	1954		II			Neg.	Mongol	M	3 da.
31	Hejlt and Wegelius[23]	1956					Neg.	Jaundice; Coombs test was negative	M	2 wk.
33	Shindo[25]	1956		I			Neg.	First examination on seventeenth day	F	17 da.
34	Shindo[25]	1956					Neg.	First examination on tenth day	?	10 da.
38	Krivit and Good[27]	1956					Neg.	First examination on third day; jaundice	M	3 da.
39	Krivit and Good[27]	1956	21				Neg.	Skin rash on third day	M	3 da.
45	Lee[29]	1957	39	VI			Neg.	Mongol; congenital heart disease unit; *Staph. aureus*	M	4 da.
37	Farber	Krivit and Good[27]						*Staph. aureus*		
41	Smith	Krivit and Good[27]						*Staph. aureus*		
42	Wolf	Krivit and Good[27]						*Staph. aureus*		
43	Andrea	Krivit and Good[27]						*Staph. aureus*		
44	Des Forges	Krivit and Good[27]								

*+ = slight; ++ = moderate; +++ = severe; ++++ = very severe; 0 = not present.

During the Neonatal Period; Summary of Case Reports

Age at death	Type of leukemia	Jaundice	Purpura	Melena	Hepato- and splenomegaly	Skin nodules	Hgb. (gm. %)	RBC	WBC	Platelets	Blasts (%)	Post-mortem
3 wk.	Lymphatic (?)	0	+*	+	++++ Kidney enlarged	0	35	2.00	115,000	6,000	95	Yes
7½ wk.	Myeloid	0	Late	0	+++	0	42	2.50	97,000	16,000	24	Spleen only
		0	+	0	+++	+						
5 wk.	Micromyeloblastic	0	+++	+	++	0	35	1.77	855,000		30	Yes
42 da.	Lymphocytic	0	++	0	0	0	Not examined at birth 8	2.8	23,000	Red	3	Yes
12 da.	Myeloblastic	0	++	0	00	0	19 10	5.20 2.80	59,000 147,000		Myelocytes 64; myeloblast 18	Yes
5 da.	Myeloid	0	0 +++	0 0	0 +++	0	14	4.19	270,000	Red	90	Yes
6 wk.	Myeloblastic	+	+	0	+++	0	3.5		448,000	Red	Atypical blasts	Yes
3 wk.	Myeloblastic	0	++	0	++	0	14.2	5.68	99,000	74,000	72	Yes
2 mo.	Myeloid	0	+	0	+	0	48	2.68	570,000		35 Myelocytes	Yes
2 mo.	Lymphatic	0	+++	0	++++	0			130,000		34	Yes
Living	Leukemic reticulosis	0	+	0	0	0	17		32,000		25	
16 da.	Stem cell	0	+	0	++	0	18			20,000	In bone marrow	
											Stem cells 26	
	Myelogenous										67 Lymphocytes	
	Myelogenous											Yes
	Stem cell											
	Myelogenous											

adrenals, pancreas, and liver; the bone marrow was replaced by blasts, and the erythroid elements were present in low percentage. Two other cases (19 and 25) developed complicating staphylococcal infections.

were invariably those who exhibited mild signs of disease in the newborn period.

In 6 cases (see Table IV) recorded as congenital leukemia, the first symptoms developed between the third and sixth week of life, and the con-

TABLE IV. CASES OF LEUKEMIA WITH ONSET OF SYMPTOMS

CASE	AUTHOR AND REFERENCES	YEAR REPORTED	MATERNAL HISTORY					CLINICAL NOTE	SEX	AGE FIRST SYMPTOM WAS NOTED
			AGE	PARA	THREATENED ABORTION	RADIATION	SEROLOGY			
7	Kornmann[8]	1934					Neg.	Icterus neonatorum	F	6 wk.
9	Kelsey and Anderson[1]	1939		I				Apparently normal at birth	M	21 da.
11	Poncher[12]	1942	(Father's and mother's marrow were examined by bone aspiration)				Neg.	1st examination at 6 wk.; hepato- and splenomegaly	M	?
12	Cross[13]	1944		I			Neg.	Melena at age 5 da.; leukemia diagnosed at 3 mo.; intraventricular septal defect	M	5 da.
13	Cross[13]	1944		I			Neg.	Melena; crusted skin lesions; "chronic, myeloid leukemia"	F	?
15	Keith[15] Potter[20]	1945 1946					?	Petechiae since age 3 weeks; leukemia diagnosed at 3 months	F	3 wk.

*+ = slight; ++ = moderate; +++ = severe; ++++ = very severe; 0 = not present.

Seven of the cases in this group were in mongol infants who were included in Krivit's and Good's[27] survey for the incidence of the simultaneous occurrence of leukemia and mongolism. Details of cases 37 and 41 to 44 have not yet been recorded in the literature, and it is possible that some of these cases exhibited signs of the disease at birth and should be listed in Table I. In case 39 significant leukemia was detected by a careful blood study in a newborn mongoloid infant who showed no other signs of the disease. The duration of life ranged from 5 days to 2 months in this group of infants. Infants who survived the longest

genital nature of the disease was accepted by the reviewers on the basis of extensive infiltration of the organs which appeared to be incompatible with the duration of the disease after birth. No case had been observed from birth to the onset of the disease. Three cases (12, 13, and 15) had shown hemorrhagic signs in the neonatal period, and the other 3 cases exhibited no symptom of the disease until after 3 weeks of life. All of the cases were diagnosed as myelogenous leukemia, and all the infants exhibited hepato- and splenomegaly and purpura at the time of hospital admission. Anemia was a constant finding. A hyperleukocytosis was

present in all the infants; the percentage of myeloblasts was variable but myeloblasts, promyelocytes, and myelocytes dominated the peripheral blood smears. The duration of life varied from 3 weeks to 3½ months in 5 of the 6 cases.

that erythropoietic factors provided by the placental circulation provide a stimulus to fetal erythropoiesis which is lost with the advent of extrauterine life. The presence of erythropoietin in cord blood is known, but the role

WITHIN 3 TO 6 WEEKS OF LIFE: SUMMARY OF CASE REPORTS

AGE AT DEATH	TYPE OF LEUKEMIA	JAUNDICE	PURPURA	MELENA	HEPATO- AND SPLENOMEGALY	SKIN NODULES	HGB. (GM. %)	RBC	WBC	PLATELETS	BLASTS (%)	POST-MORTEM
8 wk.	Myeloid	+*	+	0	+++	0	30	1.50	36,000	—	49	Yes
3 da.	Myelocytic	0	++++	0	++++	0	6.5	2.00	223,000	111,000	22 myeloblasts 68 myeloblasts	Yes
6 wk.	Chronic myeloid	0	+	0	++	0	7.6	2.45	195,000		20 myeloblasts 23 monoblasts	
3½ mo.	Myelocytic	0	+	+	+	0	55	2.25	112,000		?	Yes
3½ mo.	Myelocytic	0	+	+	++	0	5.9	2.27	23,000	72,000		Yes
15 mo.	Chronic myeloid				+++	0	8.4	2.90	29,000	73,000	7 myelocytes 2 metamyelocytes	Yes

DISCUSSION

The true congenital nature of the disease is undeniable only in those cases exhibiting signs of the disease at birth or within the first few days of life. The intrauterine disease produces widespread systemic infiltration and tumor-like collections of immature blood cells throughout the body, which appear to be tolerated better during intrauterine life than after birth. The absence of anemia at birth and the rapid fall in the hemoglobin and erythrocyte levels in the early neonatal period suggests of other maternal factors and the immunologic behavior of the fetus which determine the reaction of the infant to the disease is speculative and the subject of active research in the cancer field today.

In the group of infants who develop signs of leukemia within the first few days of life, it also appears that some maternal factor may have suppressed the intrauterine disease. However, it is debatable whether a disease which is not recognizable at birth and which develops after the first week of life is truly congenital

rather than acquired even though the degree of organ involvement may be extensive enough to persuade one that it must have developed in intrauterine life.

In the third group of infants, the evidence for congenital leukemia was based mainly in the history of a hemorrhagic symptom during the neonatal period and a failure to gain normally until the diagnosis was established at age of 3 to 6 weeks; although the diagnosis of leukemia is unquestionable in these cases, it would seem that an acceptance of the congenital nature of the disease must await further enlightment regarding its etiology.

It is significant that no case of congenital leukemia has been reported in a newborn infant whose mother suffered from leukemia during pregnancy. Erf[31] reviewed 100 cases of leukemia and lymphosarcoma complicated by pregnancy and commented upon the absence of leukemia in the offspring, and additional reports by Harris[32] and others[33-35] bring the total to 129 cases. The high fetal mortality rate in cases of maternal leukemia are well known, and Bierman and his colleagues[35] were able to find only 50 cases in which the pregnancy resulted in the birth of a normal infant. Whether the fetal loss is due to the effects of the grave systemic disease in the mother, or to the drugs employed for the control of her leukemia, or to the effect of a transmissible leukemogenic agent are questions of active inquiry in leukemia research.

Cramblett and his co-workers[36] recently reported the occurrence of acute lymphatic leukemia in a 9-month-old infant whose mother developed unrecognized symptoms of leukemia in the seventh month of pregnancy; the diagnosis of acute lymphatic leukemia was made on the eighth puerperal day, and she died two months later. The infant had shown no signs of illness in the newborn period when a normal hemogram was obtained, and he continued well until the age of 9 months when he suddenly developed anemia, signs of hepato- and splenomegaly, and a leukemic leukocytosis. The rarity of this occurrence is apparent since it is the only case of leukemia reported to have developed as the product of a pregnancy complicated by leukemia. While the single occurrence of leukemia in a mother and child may be explained by chance alone, the possibility of the transplacental transmission of a leukemogenic agent is suggested.

The virus theory of leukemia has gained recent support from the experimental leukemias of fowls and rodents. The virus nature of avian leukosis, which was suggested by the work of Engelbreth-Holm and Frederiksen,[37] has been confirmed in several laboratories. The virus etiology of mouse leukemia is still unproved, although investigations in this field have been stimulated by the observations of Gross.[38] He demonstrated that tissue extracts and cell-free filtrate of AK leukemic mice inoculated into the low-incidence C_3H strain of mice when they are 1 day old induced leukemia in a high percentage of the mice when they reached the age of 9 to 12 months and that the induced leukemia was transmitted to the offspring of the inoculated animals.[38a] Schwartz and his colleagues[39] reported an accelerating factor presumably due to virus in brain filtrates of AKr mice which hastens the onset of spontaneous leukemia in both the high-incidence AK strain and in the

low-incidence C_3H strain. More recently, a similar accelerating factor was reported in filtrates of patients with human leukemia.[39a] These experiments have not been confirmed in other laboratories as yet. The first report of the isolation of a virus which induces leukemia in mice was reported by Friend,[40] who recovered a leukemogenic virus through several passages in mice by cell-free filtrates of leukemic tissues.

The importance of maternal factors of probable etiological significance cannot be evaluated from the paucity of data provided by the case reports of congenital leukemia available in the literature. Aside from the absence of maternal syphilis, which is stated in all of the cases, there is no consistent inquiry or record regarding the maternal age, diet, infectious illnesses, exposure to radiation by pelvimetry during pregnancy, or threatened abortion. An apparent statistical correlation between the occurrence of mongolism and congenital leukemia is suggested by the report by Krivit and Good,[27] in which 39 cases of mongolism associated with leukemia were collected, and in 10 of the 39 mongoloid children, the leukemia was diagnosed in the newborn period. The correlation is further suggested by the fact that of 45 cases of congenital leukemia reported, 10 occurred in mongoloid babies. It seems highly probable that this correlation is falsely high since it is likely that the effort to collect the cases of congenital leukemia associated with mongolism has stimulated a reporting of these cases while all of the cases of congenital leukemia may not have been reported in the same time period. However, the statistical correlation between mongolism and leukemia indicates that maternal factors associated with mongolism may also be effective against the developing hematopoietic system. Maternal age and intrauterine bleeding in the early months of pregnancy appear to be important factors in mongolism, and further information on these points is desirable in the cases of congenital leukemia.

The effect of radiation of the fetus on the occurrence of intrauterine leukemia requires further inquiry. Although it is clear that radiation must account for only a small fraction of the cases of childhood leukemia, the observation of Stewart and associates[41] which suggested that irradiation of the fetus in utero increases the risk of the disease in childhood is highly significant; 13.7 per cent of the mothers of leukemic children as compared with 7 per cent of the mothers of nonleukemic children, who served as controls in this study, had been subjected to pelvimetry. In a similar study of children with various malignancies, Manning[42] found that a higher percentage of the mothers of leukemic children had received irradiation prior to pregnancy than mothers of healthy children. Such evidence supports the contention that intrauterine irradiation during the early weeks of fetal life may predispose the hematopoietic system to leukemic transformation, and, although the correlation is an indirect one, further information would seem desirable, particularly relating to the congenital leukemias.

Differential Diagnosis.—The differential diagnosis of leukemia in the newborn infant includes the consideration of infections due to pyogenic

organisms and inclusion body disease, syphilis, erythroblastosis fetalis, thrombocytopenic purpura, and aplastic anemia.

Septicemia.—In the newborn infant, septicemia offers the most serious problem in the differential diagnosis of leukemia. *Staphylococcus aureus* is the most common organism productive of a severe leukemoid reaction, although any of the pyogenic organisms, streptococcus, *Escherichia coli*, bacillus, proteus, pseudomonas, and other gram-negative bacilli may be the causative agent. If the infection is acquired in utero, clinical signs of hepato- and splenomegaly, maceration of the skin, purpura and hemorrhages into the mucous membranes and serous surfaces, and a severe degree of leukocytosis may be present at birth. When the infection is acquired after birth, these signs may develop rapidly. The leukemoid reaction to infection is an increase in the number of cells of the granulocytic series, including the transition forms from the promyelocyte stage through the various maturation forms to the segmented leukocytes. The characteristic gap or hiatus leukemicus between the predominating number of blast forms and the mature granulocytes is lacking in the leukemoid response. Anemia of a normochromic type develops rapidly, and a thrombocytopenia is often present. Jaundice is a common symptom. However, it is more often due to a hepatitis than to a hemolytic effect of the sepsis. The clinical diagnosis is established by the detection of the causative organism by blood culture or by the localization of the infection as pneumonia, osteomyelitis, or soft-tissue abscess. The pathologic findings may reveal the route as well as the nature of the infection. Although the cellular reaction may be extreme, the diffuse infiltration with immature cells, typical of leukemia, is lacking. The simultaneous occurrence of leukemia and septicemia during the newborn period presents so difficult a problem in the evaluation of the hematologic and pathologic findings that a positive diagnosis of leukemia may be untenable unless the pathologic evidence of organ infiltration with cells identifiable as blasts is well established.

Inclusion Body Disease.—Inclusion body disease in the premature and newborn infant is regularly manifested by a purpuric rash, hepato- and splenomegaly, and severe jaundice[43-44] and clinically resembles severe erythroblastosis rather than congenital leukemia. The hematologic findings are those of a leukemoid hyperleukocytosis and a secondary type of thrombocytopenia and erythroblastemia.

Congenital Syphilis.—Congenital syphilis which is severe enough to produce a leukemoid hematologic reaction in the newborn period invariably presents clinical signs of jaundice, hepato- and splenomegaly, skin manifestations, or the x-ray signs of periostitis, and the serological evidence of disease in the mother.

Erythroblastosis Fetalis.—This may resemble congenital leukemia prior to the onset of jaundice, and several of the cases reported as leukemia in the older literature have been identified as hemolytic disease of the newborn infant. While the mild cases of erythroblastosis present no signs suggestive of leukemia, the infant who

is severely affected with Rh erythroblastosis at birth shows a marked degree of hepato- and splenomegaly, thrombocytopenia, and leukocytosis of a leukemoid type. The leukocytosis, anemia, and the presence of myelocytes and even myeloblasts in the peripheral blood are indications of an excessive hematopoiesis stimulated by blood destruction. The onset of deepening jaundice, which becomes evident within the first few hours of life and which may be anticipated by a hyperbilirubinuria in the cord blood, reveals the hemolytic nature of this disease. The characteristic and regular finding of large numbers of erythroblasts in the peripheral blood excludes the diagnosis of leukemia, and the demonstration of the circulating Rh antibody by the Coombs test establishes the diagnosis of the isoimmune disease.

Congenital Thrombocytopenic Purpura.—The hemorrhagic signs are commonly present at birth. Purpuric skin manifestations may resemble the nodules of leukemia; however, the lesions are not raised, although ecchymoses may produce a diffuse swelling in the subcutaneous tissue. The purpuric skin lesions do not exhibit a rubbery consistency characteristic of nodules. The hemorrhagic signs vary widely in different cases from minimal signs of a petechial rash to massive purpuric manifestations and bleeding from mucous membranes. The spleen is usually not significantly enlarged, and hepatomegaly and lymphadenopathy are not present. The blood findings reveal an increased bleeding time, a reduction in the platelet count, and a moderate leukocytosis with no unusual elevation in the number of myelocytes or myeloblasts. Platelet agglutinins may or may not be demonstrable in the infant's serum or in the mother's serum. An anemia is absent unless hemorrhage has occurred. The prothrombin and the clotting time are within the normal range, although clot retraction is poor.

Aplastic Anemia.—Aplastic anemia of the Fanconi type, associated with purpura and a hypoplastic anemia, has been confused with congenital leukemia. In this type of aplastic anemia, skeletal deformities, particularly absence of the radii, are characteristic. The superficial resemblance presented by purpuric signs are due to the thrombocytopenia. Hepatomegaly and splenomegaly are absent and the marrow is hypoplastic.

TREATMENT

There is no effective treatment of leukemia in the newborn infant. As anemia and a progressive thrombocytopenia develop, supportive transfusions with platelet-containing fractions of blood are of temporary value. There is no case report of an effective control of the disease in the newborn infant with the antileukemic drugs such as the antifolic compounds or the purine antagonist drugs.

REFERENCES

1. Kelsey, W. M., and Anderson, D. H.: Congenital Leukemia, Am. J. Dis. Child. **58**: 1268, 1939.
2. Smith, L. W.: A Case of Acute Leukemia in an Infant, Am. J. Dis. Child. **21**: 163, 1921.
3. Koch, M. I.: Zur Frage du kongenitalen Leukämie, Zentralbl. Allg. Path. **33**: 7, 1922.
4. Stransky, E.: Beiträge zur klinischen Hämatologie im Säuglingsal, Monatschr. f. Kinderh. **29**: 654, 1925.
5. Bungeler, W.: Angeborene Leukämie, Frankfurt. Ztschr. f. Path. **41**: 257, 1931.
6. White, P. J., and Burns, E. L.: Fatal Acute Lymphoblastic Leukemia With

Great Enlargement of Kidneys in Infant 3 Weeks Old, Am. J. Dis. Child. **41**: 866, 1931.
7. Giblin, J.: A Case of Myelogenous Leukemia Occurring in Infant 5 Weeks Old, Arch. Pediat. **50**: 662, 1933.
8. Kornmann, V.: Zeitrag zur frühkindlichen und angeborenen myeloischen Leukämie, Ztschr. f. Kinderh. **56**: 440, 1934.
9. Cooke, J. V.: Personal Communication to Kelsey and Anderson.[1]
10. Bernhard, W. G., Gore, I., and Kilby, R. A.: Congenital Leukemia, Blood **6**: 990, 1951.
11. Morrison, M., Samwick, A. A., and Rubinstein, R. I.: Congenital Leukemia With Chloroma, Am. J. Dis. Child. **58**: 332, 1939.
12. Poncher, H. G., Weir, H. F., and Limarzi, L. R.: Chronic Myelogenous Leucemia in Early Infancy; Case Report, J. PEDIAT. **21**: 73, 1942.
13. Cross, F. S.: Congenital Leukemia, Report of 2 Cases, J. PEDIAT. **24**: 191, 1944.
14. Hamme, B.: Ein Fall von kongenitaler myeloischer Leukämie, Acta paediat. **31**: 330, 1944.
15. Keith, H. M.: Chronic Myelogenous Leukemia in Infancy, Am. J. Dis. Child. **69**: 366, 1945.
16. Houstek, J., and Brachfeldova, J.: Leucémie congénitale, Ann. Paediat. **172**: 98, 1949.
17. Heen, R. C.: Congenital Lymphatic Leukemia, Am. J. Dis. Child. **80**: 800, 1950.
18. Taylor, F. M., and Geppert, L. J.: Congenital Myelogenous Leukemia, Am. J. Dis. Child. **80**: 417, 1950.
19. Casilli, A. R., Rumsey, W. L., and Satulsky, E. M.: Acute Neonatal Myeloblastic Leukemia, A. M. A. Am. J. Dis. Child. **83**: 788, 1952.
20. Potter, E.: Pathology of Fetus and Newborn, Chicago, 1952, Year Book Publishers, p. 550.
21. O'Connor, R. E., McKey, R. W., and Smith, J.: Congenital Leukemia, Am. J. Dis. Child. **88**: 740, 1954.
22. Reimann, D. L., Clemmens, R. L., and Pillsbury, W. A.: Congenital Acute Leukemia; Skin Nodules, First Sign, J. PEDIAT. **46**: 415, 1955.
23. Hjelt, L., and Wegelius, R.: Congenital Leukemia, Ann. paediat. Fenniae **2**: 206, 1956.
24. Fischer, S.: Leucämia congenita, Ugesk. Laeger **118**: 445, 1956.
25. Shindo, M.: A Case of Congenital Leukemia (6th Case Reported in Japan), Tohoku J. Exper. Med. **66**: 307, 1957.
26. Strenger, W.: Angeborene Leukämie, Krebsarzt **11**: 293, 1956.
27. Krivit, W., and Good, R. A.: Simultaneous Occurrence of Mongolism and Leukemia, A. M. A. J. Dis. Child. **94**: 289, 1957.
28. Schunk, G. J., and Lehman, W. L.: Mongolism and Congenital Leukemia, J. A. M. A. **155**: 250, 1954.
29. Lee, Chi Lao: Congenital Leukemia Associated With Mongolism, J. PEDIAT. **51**: 303, 1957.
30. Fortina, A., and Petrocini, S.: Contributo allo studio delle manifestazioni cutanee nelle leucemie dell'infanzia, Pediatria **61**: 199, 1953.
31. Erf, L. A.: Leukemia (Summary of 100 Cases) and Lymphosarcoma Complicated by Pregnancy, Am. J. Clin. Path. **17**: 268, 1947.
32. Harris, L. J.: Leukaemia and Pregnancy Canad. M. A. J. **68**: 234, 1953.
33. Morgan, J. E., and Reyes, C. T.: Association of Acute Lymphatic Leukemia and Pregnancy, Obst. & Gynec. **8**: 642, 1956.
34. Gillim, D. L.: Leukemia and Pregnancy, Am. J. Obst. & Gynec. **70**: 1047, 1955.
35. Bierman, H. R., Aggeler, P. M., Thelander, H., Kelly, K. H., and Cordes, F. L.: Leukemia and Pregnancy, J. A. M. A. **161**: 220, 1956.
36. Cramblett, G., Freedman, J. L., and Najjar, Samir: New England J. Med. **259**: 727, 1958.
37. Engelbret-Holm, J., and Frederiksen, O.: Transmission of Mouse Leukosis to Healthy Animals by Means of a Cell Free Substance, Acta path. et microbiol. scandinav. Suppl. **3**: 145, 1938.
38. Gross, L.: Susceptibility of Suckling-infant, and Resistance of Adult, Mice of C_3H and C_57 Lines to Innoculation With Ak Leukemia, Cancer **3**: 1073, 1950.
38a. Gross, L.: Pathogenic Properties, and "Vertical" Transmission of the Mouse Leukemic Agent, Proc. Soc. Exper. Biol. & Med. **78**: 342, 1951.
39. Schwartz, S. O., Schoolman, J. M., Szanto, P. B.: Studies in Leukemia. IV. Acceleration of Development of AKR Lymphoma by Means of a Cell Free Filtrate, Cancer Res. **16**: 559, 1956.
39a. Schwartz, S. O.: Etiology of Leukemia: A Case for the Virus Theory, Blood **11**: 1045, 1956.
40. Friend, C.: Cell-Free Transmission in Adult Swiss Mice of a Disease Having Character of a Leukemia, J. Exper. Med. **105**: 307, 1957.
41. Stewart, A., Webb, J., Hewitt, D.: A Survey of Childhood Malignancy, Brit. M. J. **1**: 1495, 1958.
42. Manning, M. D.: Conferences on Malignancies in Childhood, School of Public Health, Harvard, Boston, 1958.
43. Smith, M. G., and Vellios, F.: Inclusion Disease or Generalized Salivary Gland Virus Infection, Arch. Path. **50**: 862, 1950.
44. Medearis, D. N.: Cystomegalic Inclusion Disease, Pediatrics **19**: 467, 1957.

Clinical Pathological Conference

The Children's Medical Center
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Sidney Farber, *Editor*
John M. Craig, *Assistant Editor*

THIS 3 3/12-year-old boy was well until 6 months before his hospital entry, when he began to run a uniform afternoon fever to 101° F. Soon he began to have nosebleeds, two to three times each week; medical examination revealed a bleeding point in the right Hesselbach's triangle in an otherwise healthy child. A routine blood examination, however, gave a hemoglobin value of 11.4 Gm. per cent, a leukocyte count of 18,000, with lymphocytes 34, polymorphonuclear cells 8, and eosinophilic leukocytes 58 per cent. The high leukocyte and eosinophil count persisted. After the onset of symptoms, he was admitted to another hospital, with occasional vomiting and a feeling of abdominal fullness. His physical examination was negative. He had a few days of fever in the hospital, attributed to an upper respiratory infection. No definite diagnosis was established. Over the next few weeks he had periodic swelling of the dorsa of the feet, the skin of these areas on one occasion developing a peculiar macular blotching.

The child was born out of wedlock, weighing 8 pounds at birth. He lived with his mother until the age of 1½ years, but no details of his health are known. He was then placed with foster parents until he was 2½ years old. These people had several pet dogs. While living with the family, the patient had two bouts of pneumonia at 6-month intervals. He was treated with penicillin but was not hospitalized. Urticaria developed after penicillin administration on one of these occasions. His prospective adoptive parents, with whom he now lives, have a cat but no dog.

On admission he was alert and active. He weighed 31½ pounds and measured 35½ inches. These figures were below the twenty-fifth and third percentile, respectively, for his age. His head circumference was 53 cm. and abdominal circumference 99 centimeters. The vital signs were: temperature 100° F., blood pressure 98 mm. Hg systolic and 54 mm. Hg diastolic, pulse 100 per minute, and respirations 24 per minute. His tonsils were large but not injected. After examination, the heart and lungs were found to be normal. The abdomen was distended, and the liver was palpable 4.0 cm. below the costal margin; it was firm and smooth, with a sharp edge, and was questionably

tender. Neither the spleen nor the kidneys were palpated. Shifting dullness but no fluid wave was elicited in the abdomen. Bowel sounds were normally active. No jaundice, rash, or edema was present.

The initial laboratory examination gave a hemoglobin level of 12.0 Gm. per cent. The leukocytes numbered 22.0 thousand per cubic millimeter. The differential smear contained normal red cells, adequate platelets, and polymorphonuclear cells 27, lymphocytes 24, monocytes 1, basophils, 1, and eosinophils 47 per cent. The corrected sedimentation rate was 27 mm. per hour. The urine was normal. The serum protein levels were: total 7.8, albumin 4.6, and globulin 3.2 Gm. per cent. The gamma globulin level was 1.25 Gm. per cent. The serum electrolytes were: sodium 137, potassium 3.7, and chlorides 102 mEq. per liter. The skin tuberculin reaction, trichinella skin test, a blood culture, liver flocculations, C-reactive protein, and antistreptolysin titers were all negative. The child's blood was determined to be group B, and his anti-A hemagglutination titer was 1:512. The stools were negative for blood, ova, or parasites. The electrocardiogram was normal, and roentgenograms of the skull and chest as well as a barium swallow for varices were all negative.

The hospital course was characterized by a low-grade fever and persistent eosinophilia. One week after entry, an operation was performed.

DISCUSSION

DR. FRANKLIN A. NEVA.*—We have, in summary, a 3-year-old boy who is not too sick but who is running a persistent leukocytosis and a high percentage of eosinophils, with a moderately enlarged liver, some free fluid in the abdomen, and an elevation of the serum gammaglobulin fraction. We might look, at this point, at the x-rays. The lungs, particularly, would interest me.

DR. EDWARD B. D. NEUHAUSER.—Dr. Neva, we have two films of the chest taken on the same day. Even in retrospect, there is nothing that I can define as a Löffler's type of infiltration or any abnormality of the heart. The liver is about top normal in size, but there is no evidence of enlargement of the spleen. A barium swallow for esophageal varices was also negative.

DR. NEVA.—A child with asthma might show an eosinophilia that would last for some time, but the only respiratory difficulty experienced by this child was two bouts of pneumonia.

The presence of an enlarged liver, leukocytosis, eosinophilia, and elevated globulins suggests a generalized process. Eosinophilic leukemia might be considered. No adenopathy was present, normal cells were apparently present in the blood smear, and the child was not seriously ill. On the basis of these facts and the statistical unlikelihood of a chronic myelogenous leukemia in a child of this age, I think we can dismiss this possibility.

Dermatomyositis and polyarteritis nodosa may be associated with eosinophilia, but there is little in this history or findings to suggest such a diagnosis.

Eosinophilia suggests a parasitic disease, particularly where there is invasion of the tissues by the parasite,

*Dr. Neva is Associate Professor of Tropical Public Health, Harvard School of Public Health.

although intestinal worm infestation may give rise to a low degree of blood eosinophilia. A skin test for trichinosis was done. Trichinosis, in a 3-year-old child, would be quite unusual because young children rarely are in a position to eat uncooked pork, either by sampling pork if sausages are made at home or by eating out in restaurants.

The stool was negative for ova and parasites, and in this connection it is worth pointing out that a number of parasitic infections can be present without evidence of their presence in the stool. In trichinosis, for example, although worms are present in the intestine during one stage of the illness, the male worms pass out very quickly in the stool and the females burrow into the submucosa of the bowel so that one can rarely demonstrate such parasites in the stool. Infection with certain blood parasites may be associated with an eosinophilia, such as with filariasis; even schistosomiasis may occur without evidence in the stool. Even though the parasite is an intestinal worm in one of its hosts, the major disease process in man may be due to infection with the larval form of the parasite, such as in echinococcal disease or as in cysticercosis caused by the the pork tapeworm, *Taenia solium*. Both of these may develop with no evidence of a worm in the intestinal tract. Even with such common parasites as the hookworm, there may be manifestations due to the larval form of the parasite. This is seen with larvae of hookworms not ordinarily infecting man, such as those of the cat and dog, which burrow into the skin and cause "creeping eruption."

Prior to January, 1952, I think this case would have been classified with those described by Zuelzer and Apt[1] under the term "chronic eosinophilia associated with visceral granulomatosis." This syndrome, usually appearing in children, consists of hepatosplenomegaly and chronic eosinophilia and the presence of granulomatous lesions in the liver. In 1952 Beaver and his associates[2] at Tulane showed that similar findings were associated in infections with the larval forms of dog and cat ascaris; this disease was named "visceral larva migrans." It is usually seen in toddlers less than 4 years of age who have access to contaminated soil containing the embryonated eggs of *Toxocara canis*. If they ingest the dirt, the larvae will hatch out and migrate through the viscera; they may reach the lungs and produce a pneumonitis, particularly if the infection is heavy enough, but the main lesions that result are the granulomata of the liver and other viscera.

As we look back over the features of this case, it fits well with the syndrome of visceral larva migrans. There was a history of exposure to dogs. It would be nice to know the age and sex of these dogs because it has been shown that male dogs are much more likely to harbor the dog ascaris than females and both sexes tend to lose the infection as they get older. Infection of dogs with the parasite is common in this country, the over-all incidence being about 20 per cent. We do not know whether this child was a dirt eater. His two bouts of pneumonitis in the 6 months prior to his admission here were probably related to larvae reaching the lungs.

Many cases of this syndrome have been recognized here at The Children's Hospital. Heiner and Kevy[3]

described three typical but unproved cases from this hospital in which they found very high anti-A and anti-B blood group hemagglutinin in titers. I do not know of any further work on this interesting relationship, although several years ago Oliver-Gonzalez and Gonzalez[4, 5] pointed out that various bacteria and parasites would absorb both the heterophil antibodies and A-2 agglutinins in human sera.

Heiner and Kevy also demonstrated evidence of specific reactions by gel-diffusion techniques in the sera of their patients against toxocara antigen prepared from the adult worms. However, because of cross reactions with a variety of other antigens, interpretation of these results may be difficult.

The definitive diagnosis of the specific agent in visceral larva migrans is difficult to make and can be done only by demonstrating the larvae in the tissues and by reconstructing the morphology of the organism by serial sections. Usually only granulomatous lesions are found and not the larvae.

Another parasite, a h e l m i n t h, which may produce a picture very similar to visceral larva migrans is *Capillaria hepatica*. A few cases of this disease have been described in humans in this country. The parasite lays its eggs in the liver, with considerable liver damage resulting, and the disease is usually more severe than that of visceral larva migrans. The infection is usually transmitted among rodents by their ingesting the carcass of an infected animal. The way in which the infection is acquired in humans is not completely understood, but it is possible that the eggs might pass through the gastrointestinal tract of a rodent and be ingested from the feces.

I assume that the operation was a laparotomy and liver biopsy. I will be very interested to know if a specific diagnosis was made in this patient.

DR. CRAIG.—Dr. Neuhauser, how do you differentiate the pulmonary infiltration of eosinophilic leukemia from Löffler's syndrome by roentgenography?

DR. NEUHAUSER.—In eosinophilic leukemia one has a diffuse evenly distributed infiltration throughout the lung, just like a miliary tuberculosis; the very few Löffler's eosinophilia that we have suspected have been isolated infiltrates, more often, I think, in the right upper lobe.

DR. CRAIG.—Dr. Diamond, would you like to say a word about eosinophilic leukemia?

DR. LOUIS K. DIAMOND.—There are certain characteristics of eosinophilic leukemia, in addition to the acute conditions observed by Dr. Neuhauser and his group, which are present in a certain number of patients, but not all. They often have marked enlargement of the spleen, like the chronic myeloid leukemia in adults, much more than we see in the acute leukemias of children or in Löffler's syndrome.

DR. CRAIG.—Have you had any experience with the heterophil antigen in this syndrome?

DR. DIAMOND.—We have had no experience with that. Dr. Neva, or Dr. Weller, is it really true that you don't see hookworm in infants? In a recent visit to Guatemala and Costa Rica, I found that some of their very young patients, even 2 or 3 months old, are affected because they have been laid

on dirt floors where the parasites invade them easily. In our community you wouldn't worry about that, of course.

DR. NEVA.—Hookworm in infancy is not common in this country, but in places where it is endemic, it may be seen in children a few months of age. When this occurs, the infection is very refractory to treatment and associated with profound anemia.

DR. JANEWAY.—What is the average course of children with eosinophilic leukemia? Is it much longer in duration than average?

DR. DIAMOND.—Yes it is, and we had one that lived two and one-half years.

DR. JANEWAY.—It is worth recalling one child, about 5 years of age, with trichinosis whose major symptoms were a large liver and eosinophilia of about 40 per cent.

A STUDENT.—Dr. Neva, could this be a hydatid cyst of the liver?

DR. NEVA.—That is a rather remote possibility. Such patients certainly develop eosinophilia if a cyst ruptures. Hydatid disease is rare in this country. This child lived up in Maine and I suppose the moose and caribou there could have hydatid disease. I would rule it out mainly on the basis of where the child lived.

DR. NEUHAUSER.—I would like to ask Dr. Weller whether we have any children with the spruce bud worm. I would also like to ask Dr. Diamond about familial eosinophilia.

DR. THOMAS H. WELLER.—I don't know how Dr. Neuhauser gets entomology into a hematologic problem! About all I know of the spruce bud worm which may be of interest is that it is now being attacked biologically through the dissemination of insect viruses by airplanes. I know of no related disease in man.

DR. DIAMOND.—Familial eosinophilia has been reported in certain families in which infections produce an eosinophilia more than they do a polymorphonuclear neutrophilia. We have had two families in which children, during periods of infection, have had as high as 20 per cent eosinophils in the peripheral blood. When we investigated the family, all the members had between 3 and 10 per cent eosinophils, even when in good health; they had been thoroughly investigated for parasites, with negative findings. It probably occurs, Dr. Neuhauser, but I don't know how to differentiate it, except by its occurrence in several members of a family.

DR. R. CANNON ELEY.—Dr. Neva, you spoke about the age relationship of trichinosis. We had here, about 15 years ago, one family of 4, the youngest 2½ years old, all of whom had trichinosis with a varying manifestation and degree of involvement. One had intercostal paralysis; another had ocular manifestations. The youngest was the least sick, and the oldest was the sickest because he had a bigger appetite and ate more of the infected pork.

DR. CRAIG.—Dr. Diamond, do you have any data as to how long the anti-A titers remain elevated in cases of visceral larval migrans?

DR. DIAMOND.—The one patient we have followed most closely still has an anti-A titer of over 100, 3 to 4 years

after his initial widespread infestation; we feel this is quite abnormal. It may be that the larvae are still present in his body and constantly act as an antigenic source. Do they ever get rid of the larvae completely, Dr. Neva?

DR. NEVA.—The larvae are quite long-lived in the experimental host. They remain alive for over a year. I think that there are cases in man where there is evidence that the larvae have probably remained alive for a year or more.

DR. DIAMOND.—Then they could remain antigenetically active?

DR. NEVA.—Yes, if that is the cause of the anti-A titer, Dr. Diamond. Is it possible to determine whether this is A-1 or A-2 antibody?

DR. DIAMOND.—It seems to be an A-1.

DR. NEVA.—Some people may be wondering, with all the remarks about antigens, whether skin tests might be used in diagnosis. So far, skin-test antigens do not seem to be very specific or very helpful in the diagnosis.

DR. MICHAEL BRAUDO.—Is there any treatment for the dogs?

DR. HANS MEIER.—Piperazine has been used, but not effectively.

DR. NEVA.—There seems to be a good deal of evidence that the infection in dogs can be transmitted in utero.

DR. CRAIG.—In the Department of Agriculture Yearbook for 1956,[6] the authors say older dogs become immune. What do you understand about "immunity" in parasitic disease?

DR. NEVA.—Immunity to parasitic infections is a complex phenomenon, the exact mechanisms of which are not yet completely understood. However, it can be demonstrated with worm infections in experimental animals which are infected repeatedly. It is much more difficult to infect the animals in subsequent exposures than when they are exposed the first time.

DR. MEIER.—Does the eosinophilia of parasite infection respond to cortisone?

DR. NEVA.—Only a few patients with visceral larva migrans have been given steroids. I don't think it is very helpful in differentiating the eosinophilia due to parasitic disease and eosinophilia due to other conditions. So far as I know, the two cases that received cortisone did not react in a dramatic fashion as far as their symptoms were concerned. It may be that the dosage of cortisone used was not large enough. In trichinosis, on the other hand, there is some evidence that treatment with cortisone results in more larvae being delivered to the tissues because of the longer life of the female producing the larvae.

DR. CRAIG.—Dr. Braudo, could you read the operative note?

DR. BRAUDO.—There is a very brief note which says that white nodules, 1 mm. in diameter, were present over the liver surface.

DR. CRAIG.—This is rather a characteristic appearance of the liver in this disease, and I don't suppose it will be a surprise that this is visceral larva migrans. We have had only one other opportunity to examine a liver

of a patient with this disease, also obtained at operation, and have never yet, until this one, been able to demonstrate the organism. Very commonly, in the portal triads one gets an appearance of a rather nonspecific lymphoid, mononuclear, and occasionally eosinophilic infiltration (Fig. 1). In other areas of the liver, one encounters granulomatous lesions with giant cells, large mononuclear cells, and scattered eosinophils which are very reminiscent, except for the presence of eosinophils, of the lesions that Dr. Jacques[7] describes in Boeck's sarcoid. He proposed a theory that perhaps some of the granulomatous lesions that one sees in Boeck's sarcoid were the end result of infestation with worms. There are other areas in which the widespread destruction of the liver with a very large area of eosinophils and mononuclears is a very loose arrangement.

Finally, we came upon a lesion in the center of which was an area of almost complete necrosis, with a cross-sectioned fragment of the worm. We did not have a chance to reconstruct this worm, although the pictures that are displayed in textbooks are quite similar to the material which we saw (Fig. 2). Dent and his colleagues[8] state that there is no proved case of *Toxocara cati* in the visceral larva migrans group. *Toxocara leonia* is another species of worm infecting older dogs.

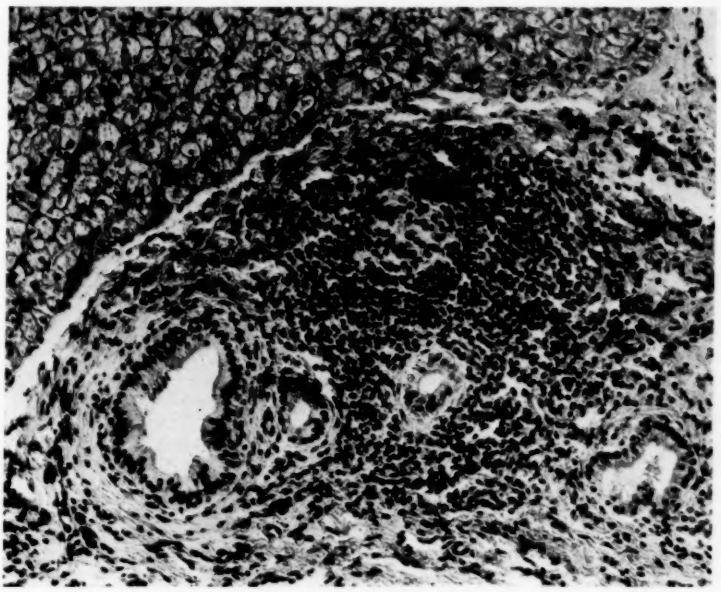

Fig. 1.—Photomicrograph of the liver. A portal triad contains a small granuloma of lymphocytes, in the center of which are a few mononuclears. (Toluidine Blue Eosin ×280.)

In one of Beaver's cases, a child with a very severe degree of visceral larva migrans received a transfusion during his hospitalization and subsequently died with serum hepatitis. They were able to isolate as many as

50 organisms per gram of liver several months after his acute episode. As many as 3 to 5 per gram were isolated from the brain, and as many as 5 per gram from the muscle. One wonders if, in some cases, a muscle biopsy might not be a very fruitful

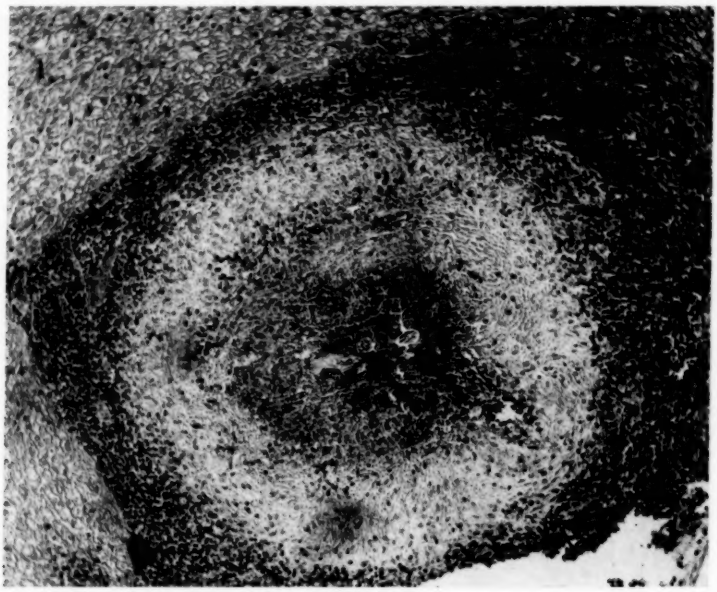

Fig. 2.—Photomicrograph of the liver. A large granuloma with an outer circle of lymphocytes, an inner rim of fibrous tissue, and a center of necrotic debris containing two cross sections of larvae at arrows. (Toluidine Blue Eosin ×170.)

and easy way to demonstrate, perhaps not the worm itself, but granulomatous lesions which would be sufficient to establish a diagnosis. This would be a much less traumatic and serious operation than doing a liver biopsy.

Dr. John Webb is here from India, and he has had considerable experience with the difficulties of tropical eosinophilia with infiltration of the lungs, and large livers.

DR. JOHN WEBB.—We have quite commonly seen children with tropical eosinophilia and a syndrome marked by respiratory symptoms, cough, asthmatic wheeziness, and a very high eosinophilia. This syndrome was first described by Frimoldt-Möller[9] in South India, and since then, this syndrome has been recognized all over India, Ceylon, Malay, and elsewhere. In the last eight months that I was in India, I learned about visceral larva migrans and, having read the papers by Beaver and seen his suggestions that tropical eosinophilia might be due to a mechanism similar to the one he described in visceral larva migrans, we began looking more critically at our cases. We found occasionally that young children, particularly, shared an enlargement of the liver as well as the striking lung infiltration. We began doing liver

biopsies in a few of them and we found this sort of nonspecific infiltration of the portal tracts to which Dr. Craig has just drawn attention. Then we did a laparotomy on a child with a typical clinical picture of tropical eosinophilia and presenting respiratory symptoms, prominent infiltrations in the x-ray, as well as a liver that was about 4 cm. below the costal margin. In that liver we found the same gross appearance as described in this case, about 12 white spots over the anterior surface, 1 to 2 mm. in diameter. A portion of tissue was taken from one of these areas and after a most wearisome search of serial sections, ultimately a portion of a nematode larva was found. Dr. Buckley of the London School of Hygiene and Tropical Medicine was unable to go further than to say that it was a nematode larva which he could not identify definitely. In 1950 left upper lobectomy was performed on a patient who was believed to have a lung abscess, but it was subsequently found that he had this tropical eosinophilia. Serial sections of this first specimen were examined and portions of larvae were found.

In Malay, where tropical eosinophilia appears to be extremely common, in a series of 150 cases, Danaraj[10] working with a technician from Beaver's laboratory showed a consistently very high titer filarial complement fixation test. This is a much higher titer than is seen in cases of infection which really are proved to be *Wuchereria bancrofti*. None of these cases of tropical eosinophilia had microfilaremia. Dr. Buckley, whom I have already mentioned, has identified a filaroid worm infecting monkeys. He brought this back to London and was interested to see if he could get it to grow in himself. He had himself infected with this organism, and then had his blood examined daily to see whether he would develop microfilaremia, but he did not. He did, however, develop a very striking eosinophilia, with a cough and respiratory symptoms. He feels that he has provided the "guinea pig" verification of a filaroid infection not normally occurring in man as being the basis for some of these cases of tropical eosinophilia in Malay. I am afraid that we haven't gotten any further than that with the investigation in India.

DR. CRAIG.—Dr. Weller, have you anything to say?

DR. WELLER.—I would like to emphasize what Dr. Webb just said—that we are opening up a whole new world of infection in man caused by parasites that normally inhabit domestic or wild animals. The work that Dr. Beaver is doing in Malay has been sponsored by the Commission on Parasitic Diseases. It was originally started because of the question—"Is tropical eosinophilia possibly synonymous with visceral larva migrans?" Clinically there are several differences. Tropical eosinophilia responds well to therapy with Hetrozan. It also responds to therapy with the arsenicals. Visceral larva migrans, which we have been talking about today, does not, and, as Dr. Webb has said, the evidence seems to be accumulating to indicate that tropical eosinophilia is probably somehow tied up with infection in man by one of these nonhuman filarial parasites. This is a situation that we may encounter here in the United States because

eases are reported, particularly from Florida and Louisiana, of a queer nodular subcutaneous lesion in man which, on section, is found by the pathologist to be something that he can just call a nematode and if he is lucky, a filarial parasite. One of the main areas in which Dr. Beaver's group is working is the identification of these worms in tissues. I think that it is an area that will be very productive, with many more findings in the next year or two.

DR. CRAIG.—He suggests, doesn't he, that the dog filaria, which is very prominent in the Southern states, may cause such a lesion?

DR. WELLER.—Well, it is possible. In some areas, 20 or 40 or even a higher percentage of dogs have dirofilaria or heart worm. It has now been shown that there are two genera that are involved. One is a filaria, mosquito transmitted, and the other is an ectoparasite. The raccoons around New Orleans have a filaria. All sorts of animals have different parasites which possibly may produce a very atypical syndrome in man.

REFERENCES

1. Zuelzer, W. W., and Apt, L.: Disseminated Visceral Lesions Associated With Extreme Eosinophilia, J. Dis. Childhood **78:** 153, 1949.
2. Beaver, P. C., Snyder, C. H., Carrera, G. M., Dent, S. H., and Lafferty, J. W.: Chronic Eosinophilia Due to Visceral Larva Migrans, Pediatrics **9:** 7, 1951.
3. Heiner, D. C., and Kevy, S. V.: Visceral Larva Migrans, New England J. Med. **254:** 629, 1956.
4. Oliver-Gonzalez, J., and Gonzalez, L. M.: Release of the A_2 Isoagglutinogen-like Substance of Infectious Organisms Into Human Blood Serum, J. Infect. Dis. **85:** 66, 1949.
5. Oliver-Gonzalez, J.: Agglutinations for Sheep Cells in Human Serums and Their Relationship to A_2 Isoagglutinogen-like Substance in Infectious Organisms, J. Infect. Dis. **90:** 44, 1952.
6. Enzie, F. D., and Price, E. W.: Internal Parasites of Dogs and Cats in Animal Diseases, The Yearbook of Agriculture, 1956, U. S. Gov. Print. Off., p. 503.
7. Jacques, W. E.: Sarcoidosis, A. M. A. Arch. Path. **53:** 558, 1952.
8. Dent, J. H., Nichols, R. L., Beaver, P. C., Carrera, G. M., and Staggers, R. J.: Visceral Larva Migrans, Am. J. Path. **32:** 777, 1956.
9. Frimoldt-Möller, C., and Barton, R. M.: Pseudotuberculous Condition Associated With Eosinophilia, Ind. Med. Gaz. **75:** 607, 1940.
10. Danaraj, T. J., da Silva, L. S., and Schacher, J. F.: The Filarial Complement Fixation in Eosinophilic Lung: Tropical Eosinophilia, Proc. Alumni Univ. Malaya **10:** 109, 1957.

Comments on Current Literature

FETOMATERNAL LEUKOCYTE INCOMPATIBILITY

THAT leukoagglutinins may develop in multitransfused patients has been recognized for a number of years, the original study of this phenomenon being stimulated by the known occurrence of autoantibodies for erythrocytes in acquired hemolytic anemia and for platelets in idiopathic thromboeytopenic purpura. The discovery by Moeschlin and Wagner[1] in 1952 of a leukoagglutinin in the serum of a patient with granulocytopenia following the administration of Pyramidon (aminopyrine) as an antipyretic led to a series of investigations concerning agglutinins for leukocytes in human sera and the possible antigens of human leukocytes. Over a period of several years, these studies resulted in the development of techniques for the detection of leukoagglutinins and in a limited survey of the disorders in which leukoagglutinins tend to occur. Attempts have been made to characterize the antibody and the leukocyte antigens and to study the leukopenia produced by passive transfer of the agglutinin. Nevertheless, the results of these studies were somewhat inconsistent, and many questions concerning the phenomenon remained unanswered. It was suggested that the therapeutic administration of blood or of drugs such as aminopyrine might not be the only stimulus for the production of leukoagglutinins. Dausset, Nenna, and Brecy[2] in 1954, and Whyte and Yee[3] in 1956 suggested an analogy to the Rh antibody formation and discussed the possibility that fetal leukocyte antigens might act as stimuli to the maternal immune mechanism. Killmann[4] also has discussed this possibility in connection with the occurrence of leukoagglutinins in collagen diseases.

Payne and her associates,[5] early interested in the correlation between blood transfusions and the development of leukoagglutinins in human sera, encountered in the course of their studies a number of persons who showed leukoagglutinins and who had no history of having received blood. At that time Payne discussed the possible significance of fetal leukocyte antigens. A recent issue of the *Journal of Clinical Investigation* carries a report by Payne and Rolfs[6] which deals with fetomaternal leukocyte incompatibility. On the basis of these studies covering a period of years, the authors feel that fetomaternal leukocyte incompatibility may result in the induction of leukoagglutinins in the mother and that "some leukocyte factors, as demonstrated by their corresponding leukoagglutinins, may be genetically transmitted to the offspring."[6] The incidence of leukoagglutinins was followed by the authors in 144 pregnant multiparous women and in a control group of 20 women; leukoagglutinins were found in 25 (17 per cent) of the 144 multiparous women. They were completely absent in the controls who had never had transfusions and who had never been pregnant. The fetus appeared to be the sole stimulus for leukoagglutinin production in 18 (68 per cent) of the 25 positive women, since these women had received neither transfusions nor any intramuscular injections of blood. While leukoagglutinins were present in some

of the women relatively early in the second trimester of pregnancy, most of the women were seen for the first time during the third trimester. The leukoagglutinin titers ranged from 1:1 to 1:128; on the whole, however, the titers were relatively low, seldom exceeding 1:8, a level comparable to that observed in the sera of multitransfused patients.[5] Autoleukoagglutinins were not present in 10 of the positive women tested. Leukoagglutinins developed in women of red cell groups A, B, and O; none were found in five women of blood group AB. In some sera, anti-red cell antibodies were observed simultaneously with antibodies for white cells. Under consideration at present is the question whether Rh-sensitized women produce leukoagglutinins more readily than do other women; to date, however, the data are not adequate to answer this question. Studies of "the nature of the leukoagglutinin leave the impression that it is an antibody separate from the factor causing L.E. cell formation, from the factor sensitizing the erythrocyte in acquired hemolytic anemia and the platelets in idiopathic thrombocytopenic purpura, and from the Rh antibody, though any one of these antibodies may or may not occur in the same sera in which leukoagglutinins are found."[5]

In an attempt to understand the stimulus for the leukoagglutinin response during pregnancy, Payne and Rolfs studied intensively the 18 women in whom the fetus appeared to be the sole stimulus, since in these patients there was no history of blood transfusions nor of intramuscular injections of blood. "If these leukoagglutinins were due to the fetal stimulation postulated, then the maternal sera should agglutinate the leukocytes of some of their respective newborn infants. If the inheritance of the leukocyte factors is analogous to that of the red cell groups, then it would be anticipated that the leukocytes of . . . (the) fathers would be agglutinated by the maternal sera containing the leukoagglutinins."[6] That is to say, some of the infants would have inherited a paternal leukocyte factor absent in the mother. In 14 family studies, maternal serum was tested against the leukocytes of the newborn infant, or the father, or both. Ten of 12 newborn infants so studied gave positive agglutination tests of this type. Examination of siblings in one of the families in which the newborn infant's tests were negative suggested that three other children in the family had inherited the father's leukocyte antigen. According to the authors' interpretation, the siblings' leukocytes could have constituted the stimuli which induced the maternal agglutinins, so that negative leukoagglutinin tests in any current newborn infant need not necessarily "invalidate the evidence for fetal stimulation." Several families studied have provided interesting and provocative results along this line, and further family studies are in progress. In six families analyzed in detail, the stimulation for leukoagglutinin production appears to have come from the fetus alone.

As the authors point out, antigenic differences in human leukocytes are known, and their existence has been demonstrated in connection with febrile transfusion reactions, even though no clear-cut types of leukocytes have been discerned. Until the present time, the main clinical significance of leukoagglutinins has been associated with their relation to transfusion reactions. Such reactions can be prevented by the use of blood from which the major portion of the leukocytes has been removed. The possibility of fetomaternal leukocyte incompatibility suggests that under these circumstances transfusion reactions on the basis of leukoagglutinins may be more serious than has been supposed in the past. Payne and Rolfs emphasize that special precautions should be taken to protect parous and pregnant women from this hazard. In one instance "a transfusion reaction associated with leukoagglutinins was observed in a woman

following her first transfusion given more than fifteen years after her last pregnancy."[6] In other recorded cases there has been evidence that leukoagglutinins persist for considerable periods of time and that their occurrence does not constitute a transitory phenomenon.

At present Payne and her associates are continuing their investigations concerning human leukocyte immunology, both with reference to transfusion reactions and with reference to the inheritance of leukocyte factors, including the possible effects of maternal leukoagglutinins on the developing fetus and the newborn infant.

RUSSELL J. BLATTNER

REFERENCES

1. Moeschlin, S., and Wagner, K.: Agranulocytosis Due to the Occurrence of Leukocyte-agglutinins (Pyramidon and Cold Agglutinins), Acta haemat. (Basel) **8**: 29, 1952.
2. Dausset, J.: Leuco-agglutinins: IV. Leuco-agglutinins and Blood Transfusion, Vox Sang. (Basel) **4**: 190, 1954.
Dausset, J., Nenna, A., and Brecy, H.: Leukoagglutinins: V. Leukoagglutinins in Chronic Idiopathic or Symptomatic Pancytopenia and in Paroxysmal Nocturnal Hemoglobinuria, Blood **9**: 696, 1954.
3. Whyte, H. M., and Yee, I. L.: Blood Groups and Agglutination of Leukocytes, Australa. Ann. Med. **5**: 214, 1956.
4. Killmann, S.-A.: Leukocyte Agglutinins in Collagen Disease, Acta rheum. scandinav. **3**: 209, 1957.
5. Payne, R.: Leukocyte Agglutinins in Human Sera, A. M. A. Arch. Int. Med. **99**: 587, 1957.
The Association of Febrile Transfusion Reactions With Leuko-agglutinins, Vox Sang. (Basel) **2**: 233, 1957.
The Significance of Leuko-agglutinins in Transfusion Practice, Presented Am. A. Blood Banks, Nov., 1957.
6. Payne, R., and Rolfs, M. R.: Fetomaternal Leukocyte Incompatibility, J. Clin. Invest. **37**: 1756, 1958.

News and Notes

The **Sociedad Jalisciense de Pediatria of Mexico** named, as officers for 1959, Dr. Antonio Martín del Campo, president; Dr. José Ceballos González, vice-president; Dr. Carlos Alberto Pelayo, treasurer; Dr. Jesús Hernández Ramírez, secretary; Dr. Julio Novon Niz, member; and Dr. Alfredo Zepeda Camarena, permanent secretary.

The **Baton Rouge Pediatric Society** announces a program by Dr. Alan Monerieff, Chief of Staff, The Hospital for Sick Children, Great Armond Street, London, England, June 16, as the Society's quarterly meeting. For further information, contact Francis M. Harris, Jr., M.D., Secretary-Treasurer, Baton Rouge Pediatric Society, 1401 North Foster Drive, Baton Rouge, La.

The Intermountain Pediatric Society announces its annual meeting which is scheduled for June 26, 27, and 28 at Sun Valley, Idaho. Dr. Robert Alway, Dr. M. E. Lahey, Dr. Ralph Platou, and Dr. Josef Warkany will speak at the meeting which is open to all physicians. Sessions are daily from 8 A.M. until noon, with round table groups from noon until 1:30 P.M. For further information, write to Intermountain Pediatric Society, 2000 South 9th East, Salt Lake City, Utah.

Diagnosis and Treatment of Thyroid Diseases is the subject of the *Fifth Annual Surgery, Radiology, Pathology Symposium* May 15 and 16, 1959, at **The University of Oklahoma Medical Center.** For further information, write to the Office of Postgraduate Education, University of Oklahoma Medical Center, 801 Northeast 13th Street, Oklahoma City 4, Okla.

The **Cook County Graduate School of Medicine** announces a 2-week course in Neuromuscular Diseases of Children with special emphasis on cerebral palsy, to be given by Dr. Meyer A. Perlstein, June 15 to 26. An intensive didactic and clinical course designed for pediatricians, orthopedists, neurologists, psychiatrists, and physiatrists interested in the care and treatment of children with neuromuscular handicaps, emphasis will be on the practical clinical aspects of treatment and rehabilitation procedures. The $250 fee will include cost of luncheons and special field trips to demonstration clinics and treatment centers.

Registration is limited. For further information, write to John W. Neal, Registrar, Cook County Graduate School of Medicine, 707 S. Wood Street, Chicago, Ill.

Applications are being received for 3 to 6 month residencies in Pediatric Rehabilitation at the **Jewish Chronic Disease Hospital,** Brooklyn, New York. Residents are given experience in the rehabilitation approach to the chronic diseases of children, including neuromuscular disturbances, congenital heart disease, and cleft palate, and to a variety of communication and psychologic handicaps. Experience is also available in cardiac catheterization and angiocardiography.

The service is a teaching unit of the State of New York Medical School. Applicants receiving their pediatric training in an accredited institution will receive 3 months of credit toward certification by the American Board of Pediatrics. Vacancies are open for July 1, 1959. The stipend is $200 a month.

Applications should be sent to Director of Pediatrics, East 49th Street and Rutland Road, Brooklyn 3, N. Y.

Fellowships in Adolescent Medicine at the **Boston Children's Hospital** are again available to properly qualified applicants. A recent grant to the hospital's adolescent unit has made it possible to offer a limited number of stipends to physicians who have had at least two years of hospital experience and who have a special interest in the medical care of adolescents. All Fellows are enrolled in The Harvard Medical School's 12-month postgraduate course, "Adolescent Medicine." Further information may be obtained by writing The Adolescent Unit, Children's Hospital, 300 Longwood Avenue, Boston 15, Mass.

The *W. T. Grant Foundation* has enabled the Children's Psychiatric Service of the **Harriet Lane Home** to offer two fellowships providing training in the psychological aspects of child medical care. These fellowships, with stipends of $6,000 per annum, will become effective on July 1, 1959. Under special circumstances, a fellowship of 6 months may be considered.

Candidates should have completed their formal pediatric training. Preference will be given to candidates interested in an academic career in pediatrics who will carry the psychological skills developed into their teaching.

Experience will be in diagnostic appraisal of the psychiatric problems of children, in short-term management of suitable cases under supervision, and in the therapeutic program of a nursery school for disturbed children. The program will be flexible enough to allow for selection of additional experience with psychosomatic problems, well-child care, public health pediatrics, etc. The program aims to prepare physicians who will be able to provide comprehensive care in pediatrics; it is not designed to train child psychiatrists.

Applications should be made to Dr. Leon Eisenberg, Associate Professor of Psychiatry and Pediatrics, Johns Hopkins Hospital, Baltimore 5, Md.

The Children's Hospital of Philadelphia announces receipt of a $100,000 grant from the Eugene Kardon Memorial Fund to further the treatment and research of cancer in children.

Dr. Stuart S. Stevenson has been appointed Professor and Chairman of the Department of Pediatrics of the recently formed **Seaton Hall College of Medicine,** Jersey City, N. J., effective March 1, 1959. Dr. Stevenson has been Research Professor of Pediatrics at the University of Pittsburgh since 1949. In addition this last year he has been Acting Medical Director of the Children's Hospital of Pittsburgh and Acting Chairman of the Pediatric Department of the Medical School.

Books

Hormone Production in Endocrine Tumors. Edited by G. E. W. Wolstenholme and Maeve O'Connor, Ciba Foundation, Colloquia on Endocrinology, vol. XII, Little, Brown & Co., Boston, 1958, 351 pages. Price $9.00.

This conference is the twelfth in a series of Colloquia on Endocrinology. It includes a valuable accumulative index to all twelve volumes. Dr. Ralph Dorfman served as chairman of the conference.

The symposium is concerned with hormone producing functional tumors of the endocrine system. Biochemical studies of tumors, which include chromatography, spectroscopy, and radioactive tracers, as well as some studies concerned with the physiology and pathology of tumors, are discussed. Twenty papers were presented at the conference and a discussion of each paper is recorded at the end of the chapter. There were twenty-nine participants.

This book would be of most value to investigators interested in experimental endocrinology but would also be of interest to the clinician because of some clinical applications which were discussed.

Hulda J. Wohltmann, M.D.

Proceedings of the Ninth Annual Conference on the Nephrotic Syndrome. Edited by Jack Metcoff, M.D., sponsored by the National Nephrosis Foundation, Inc., 143 E. 35th Street, New York, N. Y., 1958, 248 pages. Price $5.25.

The ninth conference was held at the National Institutes of Health, Bethesda, Maryland, in October, 1957. The discussion was centered around the following topics: the relationship of renal lesions to antigens and antibodies, lipoprotein metabolism, electrolyte metabolism, and evaluation of steroid therapy.

The data presented should be of value to those interested in investigation in renal disease, and the last topic, evaluation of steroid therapy, should also be of interest to the practicing pediatrician.

Hulda J. Wohltmann, M.D.

Editor's Column

TONSILLECTOMY DURING THE POLIOMYELITIS SEASON

LAST summer the question arose whether the recommended prohibition of elective tonsillectomy and adenoidectomy during the polio season should be relaxed. The question will arise again this summer, with especial reference to children who have received 3 injections of Salk vaccine, administered at appropriate intervals. The operations have been discouraged in the past during the polio season because of the increased risk of bulbar poliomyelitis[1] at a time when there is increased dissemination of poliovirus. There is ample evidence that the operation opens pathways to the entry of virus and permits its direct transmission to the central nervous system.

It has been known for a long time that poliovirus is present for several days in the pharynx of infected persons in the early stages of their illness or carrier state and that it may be found in the intestinal tract for a much longer time. There is good evidence that successful immunization with the Salk vaccine does not prevent the wide dissemination of virus.[2-4] It continues to be present in the feces and it has also been recovered from the pharynx of children who had received one or two[3, 4] or even three[5] doses of Salk vaccine.

Furthermore, the Salk vaccine, as prepared commercially, is not of uniform potency. The U. S. Public Health Service estimates its effectiveness in the prevention of paralysis at about 70 per cent.[6] During 1958, 322 cases of paralytic poliomyelitis were reported in the United States in persons who had received 3 injections of Salk vaccine.[7] Twenty deaths from poliomyelitis occurred in persons who had received 3 injections of vaccine.[7] It is impossible to know whether a particular child has been adequately immunized against poliomyelitis without performing complicated serologic tests.

These factors were considered by the Expert Committee on Poliomyelitis of the World Health Organization which met in Geneva in 1957. Its recommendations were as follows:

"Elective tonsillectomies and adenoidectomies have been forbidden in many communities during the poliomyelitis season or during epidemics of poliomyelitis because of evidence indicative of an increased risk of acquiring bulbar poliomyelitis. The mechanism here is believed by some to involve actual contamination of the nerves with poliovirus present in the pharynx. Since immunization with inactive poliovirus vaccine has been

found to have no significant effect on the excretion of poliovirus in the stools, it is possible that in vaccinated individuals with low levels of antibody there may also be no interference with multiplication of virus in the pharynx. Accordingly, it was the opinion of the committee that, at least until evidence to the contrary is accumulated, it would be advisable to continue the prohibition of elective tonsillectomies and adenoidectomies during the so-called poliomyelitis season."

Recently the opinions of eleven prominent American workers in the field of poliomyelitis were sought on the advisability of performing tonsillectomy during the polio season in children who had received 3 injections of Salk vaccine. Eight objected to the procedure, several of them calling attention to the statement of the Expert Committee. Of the three who did not specifically object, one stated: "I have been of a mind to believe that vaccination or even gamma globulin before tonsillectomy might well eliminate the risk of bulbar poliomyelitis. I realize that the information is still inadequate, but in view of the evidence that vaccination does largely prevent the paralytic disease, I have tended in this direction." Another stated that "immune individuals should be able to resist provocative effects of these procedures." The third (Dr. A. B. Sabin) felt that tonsillectomies could be performed during the polio season provided that the patient had had at least 3 injections of poliovaccine, the last dose having been administered not more than six months prior to the date of operation; otherwise, a booster dose should be given two weeks prior to the operation. He also gave this advice at a meeting of the Eastern Section of the American Laryngological, Rhinological and Otological Society in New York City on Jan. 8, 1959. The statement received wide publicity in the lay and medical press.

Since then Sabin has modified his views to require four doses of vaccine and a fifth dose two weeks prior to operation if the interval since the last dose is three months or more and if poliomyelitis is not epidemic in the community.

The weight of opinion strongly favors prohibiting the operation during the polio season even in children who have received an adequate number of injections of Salk vaccine. Such advice should be readily accepted by conscientious physicians who need not feel that they are withholding a beneficial measure. Considerable doubt has been raised about any advantages resulting from the operation. Bakwin[9] has discussed "The Tonsil-Adenoidectomy Enigma" recently in a stimulating article in this journal. Among the facts mentioned by him is one which is unfamiliar to many physicians: from 1950 to 1955, from 220 to 346 persons died each year from the direct effects of the operation in the United States.

MORRIS GREENBERG

REFERENCES

1. Siegel, M., Greenberg, M., and Magee, M. C.: Tonsillectomy and Poliomyelitis. II. Frequency of Bulbar Paralysis 1944-1949, J. PEDIAT. 38: 548, 1951.
2. Fox, J. P., Gelfand, H. M., LeBlanc, D. R., and Rowan, D. F.: The Influence of Natural and Artificially Induced Immunity on Alimentary Infections With Poliovirus, Am. J. Public Health 48: 1181, 1958.

3. Wehrle, P. F., Reichert, R., Carbonaro, O., and Portney, B.: Influence of Prior Active Immunization on the Presence of Poliomyelitis Virus in the Pharynx and Stools of Family Contacts of Patients With Paralytic Poliomyelitis, Pediatrics **21**: 353, 1958.
4. Davis, D. C., Lipson, M. J., Carver, D. H., Melnick, J. L., and Robbins, F. C.: The Degree and Duration of Poliomyelitis Virus Excretion Among Vaccinated Household Contacts of Clinical Cases of Poliomyelitis, Pediatrics **22**: 33, 1958.
5. Howe, H. A.: Personal Communication.
6. Department of Health, Education, and Welfare, Public Health Service, Communicable Disease Center: Poliomyelitis Surveillance Report, Suppl. 16, 1958.
7. Idem: Report 161, Jan. 2, 1959.
8. Expert Committee on Poliomyelitis, Second Report: World Health Organization, Technical Report Series 145, Geneva, 1958.
9. Bakwin, H.: The Tonsil-Adenoidectomy Enigma, J. PEDIAT. **52**: 339, 1958.